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NITROGEN AVAILABILITY OF GREEN MANURES

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Bureau of Plant Industry, United States Department of Agriculture

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Maintenance and restoration of humus and nitrogen in American soils is a problem of steadily increasing importance. Because of climatic conditions American soils are especially exposed to deterioration by rapid destruction of humus. Accordingly, the necessity of incorporating organic residues into the soil deserves most careful attention.

Animal manure has always held the first place in European agriculture, whereas green manure, although used since ancient times, has served more or less as a substitute. In America, as economic conditions often favor green manuring, accurate knowledge of the results to be expected from the application of this method is highly desirable.

At present, the amount of reliable data available in this direction is not very large, and opinions differ widely concerning the extent to which the green manure nitrogen may become available to succeeding crops. The investigations herein reported were made in order to secure additional information upon this important problem. They were started in 1914, and the data presented cover a period of 10 years.[1]

LITERATURE

Frequently the statement has been made that by using leguminous plants for green manure large quantities of nitrogen, equivalent to an almost seven-fold amount of nitrate, could be drawn from the air without cost. In such cases the cost of growing green manure crops has been overlooked, and furthermore, accurate tests have shown green manure nitrogen to be more or less inferior to nitrate nitrogen. There are enough valid reasons for the increased use of legumes in American agriculture to make unfounded recommendations more than superfluous.

Nitrate nitrogen is directly accessible to the roots of the cultivated plants, whereas most of the green manure nitrogen at first enters the metabolism of the soil microflora and microfauna. To what extent and how soon it will become available as plant-food depends on many variable circumstances, which are responsible for the somewhat slow and uncertain nitrogen availability of green manures. The efficiency of mineral nitrogenous fertilizers is likewise not constant, because of losses by leaching and by bacterial action. But the fluctua-

[1] The work done in the field and in the greenhouse was mainly under the supervision of Messrs. F. M. Scales and N. R. Smith. Most of the analyses and calculations have been made by Messrs. A. P. Harrison and K. S. Markley. The illustrations accompanying this report were prepared by Mr. F. L. Goll. The writer is greatly indebted to his associates and assistants for their faithful collaboration.

tions and reductions are, as a rule, greater with organic manures than with ammonia and nitrate. However, the production of carbon dioxide from decaying organic residues and their manifold influences upon biochemical processes in the soil may occasionally increase the nitrogen efficiency of organic manures beyond that of mineral fertilizers.

Experimental data published before 1910 (25) indicated that the nitrogen availability of leguminous green manures may vary between 16 and 87 per cent, and that generally a return of about 50 per cent of the total nitrogen in the manured crops is to be expected. These figures are higher than those usually obtained with animal manures, wherein the nitrogen availability was found fluctuating between 10 and 40, most frequently around 25 per cent (25). More recent observations confirm and amplify these earlier findings. A number of field tests (16, 47) showed not more than 10 to 20 per cent availability of green manure nitrogen, while more frequently 30 to 40 per cent was regained in regular farm practice (17) and approximately 30 to 50 per cent in carefully conducted trials on experimental fields (40). In America higher temperatures than in Europe, heavier rains, and periods of warm weather during winter, together with the common practice of leaving the fields bare for a considerable time of the year, even where this is not necessary for conserving soil moisture for the following crops, cause, as a rule, larger losses of nitrogen not only from organic manures but also from mineral fertilizers. Accordingly, pot experiments wherein such losses are lower or absent, produce regularly higher results (43).

The same holds true in regard to nitrification tests made under laboratory conditions. In Californian soils up to 50 per cent of the total nitrogen of green manures was converted into nitrate within three weeks; Melilotus and Cowpea (*Vigna caljang*) proved to be superior to soybean (29). Experiments made in India showed within four weeks 50 per cent nitrification of the nitrogen contained in cowpea leaves and 15 per cent from the cowpea roots (20); 50 to 60 per cent of the nitrogen of *Crotalaria juncea* became mineralized within eight weeks (19). Analogous four-month tests made with green rye and green vetch in different soils (from California and from the District of Columbia) showed almost complete transformation in the case of rye—94.7 per cent—whereas the vetch gave 73.5 per cent nitrification (49).

Such results explain the slight effect of green manuring when plants rich in water and nitrogen are plowed under while the air and soil temperatures are high and a replanting of the manured field is delayed for months. The nitrate rapidly formed is easily washed away by a few heavy rains. Large quantities of rather coarse material such as almost mature rye and oats, which offer more resistance to complete decomposition, will give better results than legumes under such conditions. Humus formation and the resulting increase in the water-holding capacity of the soil are in this case of greater influence upon the succeeding crop than the amount of nitrates derived from the green manures (6, 22).

The age at which the plants, both non-legumes (28) and legumes (3) are turned under, may or may not greatly affect their nitrogen availability. Much more nitrate is to be expected from broad-leaved plants than from cereals, and the nitrification is greatly depressed if dry stems predominate in the green manures.

The comparatively high nitrification of immature material, however, is usually more than compensated by the lower weight of the young plants. The surface growth of a crop of legumes in full bloom is as a rule twice as heavy as before blooming (31). Field experiments made in Alabama (10) with hairy vetch showed for instance an increase of nitrogen in the vines from 117 to 173 pounds and in stubble and roots from 20 to 29 pounds per acre. Data secured in Delaware (35) for crimson clover gave 56 and 108 pounds nitrogen per acre in the tops and 13 and 38 pounds in the roots. Pot experiments with soybeans (24) as well as with soybeans and cowpeas (45) gave three- to four-fold increases, which are partly due to the more favorable conditions under which the plants are grown in pot experiments.

The nitrogen availability for stubble and roots is probably always much lower than that of the surface growth with the exception of the easily decomposed root nodules, whose weight is insignificant. The statement is frequently repeated in the American literature

that about one-third of the total amount of nitrogen present in a leguminous crop is contained in stubble and roots, but it must be emphasized that such high figures are more an exception than the rule. Tests made with peas in Alabama (26) gave 58 to 227 pounds nitrogen per acre for the vines, and only 3 to 14 pounds for the roots. Experiments conducted at the same station (9, 10) with crimson clover and hairy vetch showed 120 to 173 pounds nitrogen present in the tops, and 24 to 29 pounds in the roots; the analogous figures for cowpeas were 48 and 5.68, respectively (11). Almost identical results have been recorded at the Cornell Station (8). One-tenth or less of the total nitrogen was found in the soybean, cowpea, vetch, and crimson clover roots harvested at the Delaware Experiment Station (34), whereas the red clover and alfalfa roots contained one-half or three-fourths as much nitrogen as the surface growth. The latter, however, was subnormal (55 to 70 pounds nitrogen per acre). Heavier growth of alfalfa (with 122 pounds nitrogen per acre) showed one-half of the nitrogen present in the roots, and sweet clover (with 128 pounds nitrogen) approximately one-eighth (2). In old clover and alfalfa fields occasionally very conspicuous quantities of nitrogen have been found in the roots (1, 5), but such results should not be generalized (25, p. 578). This also holds true concerning some data obtained at the Cornell Station (37) with different clovers sown in August and harvested in November of the same year, viz., in crimson clover tops, 125; in the roots, 31 pounds; in red clover tops, 63; in the roots, 40 pounds; in Mammoth clover tops, 67 pounds; in the roots, 78 pounds nitrogen per acre. In field experiments conducted in Indiana at the Purdue station for four years (48) one-seventh of the total nitrogen harvested in cowpeas, and one-ninth of that in soybeans was found in the roots. Numerous observations recorded in the German literature (31, 33) indicate likewise that clover residues may contain one-third to one-half of the total nitrogen, whereas other legumes usually do not leave more than one-eighth or one-tenth of the total nitrogen in their stubble and roots, sometimes even not more than one-twentieth (41).

These data are of fundamental importance for a correct understanding of the beneficial effect exerted by the legumes upon succeeding crops. The availability of the nitrogen in the roots will rarely surpass one-third that in the tops (21); therefore, if a leguminous crop contains 150 pounds total nitrogen of which 15 to 30 pounds remain in the stubble and roots, not more than about 2 to 5 pounds nitrogen per acre will become available to the following crop from this source. Undoubtedly no conspicuous effect can be expected from this small amount of nitrogen.

How much of the legume nitrogen has been assimilated from the air, is another point upon which widely divergent estimates are recorded in the literature. Under field conditions invariably some soil nitrogen is taken up by the roots, and it is undoubtedly a mistake to calculate all the nitrogen in a leguminous crop as a clear gain in nitrogen from the air, as is sometimes done. If a soil is originally free from root-nodule bacteria and its nitrate content is very low, well inoculated legumes will draw almost all the nitrogen they need from the air, whereas uninoculated test plants will show very little development. Actual gains of 100 pounds per acre have been observed under such conditions with hairy vetch, and 140 pounds with crimson clover in experiments made in Alabama (9). Analogous tests made with alfalfa in Illinois (18) furnished up to 90 pounds per acre, with hairy vetch in Connecticut (39) approximately 70 pounds, and with different legumes in California (30) again 90 pounds per acre. Alfalfa experiments conducted in Wisconsin (13) showed that in this case 40 per cent of the nitrogen was taken from the soil and 60 per cent from the air. Other tests indicate that from 15 to 90 per cent of the total nitrogen may have been drawn from the air (5). Since, however, most soils do not furnish more than 25 to 50 pounds nitrate nitrogen to such non-leguminous crops as are generally grown in this country, and since on the other hand an average crop of legumes, even after removal of the surface growth, does not decrease, but more often increases, the supply of total nitrogen in the soil, it seems safe to assume that at least two-thirds or three-fourths of the 100 or 150 pounds nitrogen per acre harvested in a leguminous crop has come from the air. Legumes well

adapted to the particular soil and climate will almost invariably produce crops that contain as much or even more nitrogen (17; 25, p. 667; 33), although returns of 50 or 60 pounds per acre are accepted as fair averages by some authors (42).

The cost per pound nitrogen varies according to the size of the crop and to the expenses for preparation of the soil and for seed. They have been estimated at 2 to 8 cents per pound (33) for a normal crop of about 125 pounds nitrogen per acre. With 50 per cent average nitrogen availability the use of legumes for green manuring is undoubtedly more economical than that of artificial nitrogenous fertilizers, although, as a rule, it is still more advisable to use the legumes as feed, and to return the animal manure to the field whenever this is feasible.

METHODS

The availability of green manure nitrogen depends mainly on the following three factors:

1. Quality and quantity of the green manure.
2. Time of application and quality of the soil.
3. Kinds of crops that follow after the green manuring.

Vegetation tests, therefore, are the only means of ascertaining with fair accuracy the effect exerted by these factors.

If the nitrogen availability depended merely on the quality of the green manure plants, such a determination might be possible by chemical tests. Several methods have been recommended for this purpose. The amount of nitrogen soluble in water or in water saturated with carbon dioxide has been declared to be an index of its availability (32). Occasionally 40 or 50 per cent of the total nitrogen has been found to be soluble (32, 45). This apparent agreement with the results of other availability experiments has been accepted as proof of the correctness of that assumption. But more frequently the nitrogen solubility has been found to be much lower. Very little of the soluble nitrogen is present as ammonia, about one-half of it occurs as amino acids (15). The possibility exists that these nitrogenous compounds may be directly absorbed by the manured plants (12, 23), but since they are readily attacked by numerous soil bacteria, as a rule mineralization will take place (4). The permanganate test, used for determining the available nitrogen in commercial organic fertilizers, is likewise not applicable. Its unreliability has become more and more evident (14, 38).

Nitrification tests, on the other hand, are undoubtedly better suited for securing information upon the rapidity and the extent of the mineralization of organic nitrogen. However, the results obtained are strongly influenced by the conditions under which these tests are made. Not only the quality, but especially the quantity of the organic substances which are added to the soil is of great influence; and for analytical purposes much larger quantities always must be used in nitrification experiments than are applied in farm practice. Furthermore, soils vary considerably in their nitrifying efficiency, not only from one another, but also within themselves because of seasonal and other changes. In laboratory tests, temperature, moisture, and aeration differ more or less from those regulating field nitrification. The time factor exerts its influence, too. Therefore, in general the results of nitrification tests can not be accepted as of definite value. Improvements in this direction are quite feasible, but at present the vegetation experiment undoubtedly deserves preference.

Vegetation tests in the field are of greatest practical value, but lack of uniformity in the soil, nitrogen losses by leaching, and many other disturbing factors will always impair the reliability of the results so obtained. If the land destined for experimental purposes is repeatedly tested in advance by weighing the crops grown on the different plots with uniform treatment, the selection of an unsuitable location can be prevented. The field experiments made in connection with these investigations were all based upon the outcome of such preliminary tests.

Vegetation tests in the greenhouse are free from many of the disadvantages of field tests, but the different conditions in the two tests will influence the results.

In order to ascertain the full effect of any kind of organic manure, the experiment must be of sufficient duration. As a rule, several years are necessary for the complete mineralization of nitrogen applied in this form.

If legumes are grown on the same soil wherein the manuring effect is to be tested, the results do not demonstrate exclusively the nitrogen availability of the plants incorporated in the soil. Accordingly, the plant material used for the availability experiments was grown on field plots, harvested at different times, and applied after it had been carefully dried.

Drying of green manures, which is unavoidable in such comparative experiments as were planned, has been found to cause a reduction in their efficiency especially with single crop tests (6, 19, 36, 44, 46). But if the drying is done slowly, if all losses of leaves are avoided, and if the experiments are of long duration, the final results are not seriously affected. A few comparative tests made with corn, kafir, and milo, manured with fresh and dried pea tops of the same nitrogen content gave lower weights but larger nitrogen returns from the dried green manure.

PEA VINES APPLIED	INCREASE IN FRESH WEIGHT			INCREASE IN NITROGEN EFFICIENCY		
	Corn	Kafir	Milo	Corn	Kafir	Milo
	per cent	*per cent*	*per cent*	*per cent*	*per cent*	*per cent*
Fresh	63	70	55	56	3	7
Dried	57	55	33	50	23	40

The green manures used in the pot experiments were applied in quantities of 30, 10, and 3.3 gm. dry weight, respectively. These amounts are approximately equivalent to 4½, 1½, and ½ tons air-dry substance with 225, 75, and 25 pounds nitrogen per acre, and represent a large, a medium, and a small application of green manure. The smallest quantity represents practically that usually left on the field as stubble after a legume has been cut for hay.

The various legumes had been taken from the field before the buds opened (young), when one half of the growth was in full bloom (medium), and after blooming when the pods had been partly formed (old). Ripe cowpeas with mature seed, before and after they had been exposed to frost, were included in the test.

Two different soils were used: a heavy clay subsoil of low activity and low in humus (0.103 per cent N), and a very fertile greenhouse soil of fairly high activity and well enriched with humus and nitrogen (0.143 per cent N). Phosphate and potash were adequate in both soils, and the original reaction was close to neutral. Nevertheless, in order to prevent any possible lack of lime, phosphate, and potash that might have occurred after a number of crops had been taken, alternating applications of 2 gm. monopotassium-phosphate, and 5 gm. calcium carbonate per pot were applied to every second planting.

For each series 296 galvanized pails were used. Every pot was filled with 5 pounds gravel and 20 pounds soil. Two strong glass tubes, reaching from the top down into the layer of gravel, assured proper aeration. The plan for the pot tests is shown in table 1.

After the pots had been filled with gravel and soil, in order to ascertain the uniformity of the soils a crop of unmanured buckwheat was first raised on all pails. The results of this test were quote satisfactory on the poor soil, but the rich soil, with its larger amount of available humus nitrogen, gave less consistent figures as is shown by the data for the mean probable error in tables 2 and 3. Renewed mixing of the contents of a few pails which gave exceptionally high or low crops eliminated these inequalities. Accordingly, the mean

TABLE 1

Arrangement of pot tests

POOR SOIL (NORTH HOUSE)					RICH SOIL (SOUTH HOUSE)				
Manures	N	Per pot		Pot numbers	Manures	N	Per pot		Pot numbers
		Dry	N				Dry	N	
	per cent	*gm.*	*mgm.*			*per cent*	*gm.*	*mgm.*	
None	....	....	.....	101–801	None	....	....	.....	151–851
Pea, medium	2.85	30.0	855.0	102–802	Sweet Clover, young	2.54	30.0	762.0	152–852
		10.0	285.0	103–803			10.0	254.0	153–853
		3.3	94.1	104–804			3.3	83.8	154–854
White Lupin, young	2.37	30.0	711.0	105–805	Sweet Clover, medium	2.68	30.0	804.0	155–855
		10.0	237.0	106–806			10.0	268.0	156–856
		3.3	78.2	107–807			3.3	88.4	157–857
None	....	....	.....	108–808	None	....	....	.....	158–858
White Lupin, medium	1.78	30.0	534.0	109–809	Sweet Clover, old	2.13	30.0	639.0	159–859
		10.0	178.0	110–810			10.0	213.0	160–860
		3.3	58.7	111–811			3.3	70.3	161–861
White Lupin, old	1.63	30.0	489.0	112–812	Cowpea, young	3.54	10.0	354.0	162–862
		10.0	163.0	113–813			3.3	116.8	163–863
		3.3	53.8	114–814					
None	....	....	.....	115–815	Cowpea, medium	2.71	10.0	271.0	164–864
							3.3	89.4	165–865

Hairy Vetch, young	3.27	30.0	981.0	116–816
		10.0	327.0	117–817
		3.3	107.9	118–818
Hairy Vetch, medium	2.71	30.0	813.0	119–819
		10.0	271.0	120–820
		3.3	89.4	121–821
Horse Bean, young	2.97	10.0	297.0	122–822
		3.3	98.0	123–823
None	....	....	.....	124–824
Horse Bean, old	2.91	30.0	873.0	125–825
		10.0	291.0	126–826
		3.3	96.0	127–827
Yellow Lupin, young	3.00	30.0	900.0	128–828
		10.0	300.0	129–829
		3.3	99.0	130–830
None	....	....	.....	131–831
Yellow Lupin, medium	2.01	30.0	603.0	132–832
		10.0	201.0	133–833
		3.3	66.3	134–834
Yellow Lupin, old	1.27	30.0	381.0	135–835
		10.0	127.0	136–836
		3.3	41.9	137–837
None	....	....	.....	166–866
Cowpea, old	2.18	30.0	654.0	167–867
		10.0	218.0	168–868
		3.3	71.9	169–869
Cowpea, ripe	2.00	30.0	600.0	170–870
		10.0	200.0	171–871
		3.3	66.0	172–872
None	....	....	.....	173–873
Cowpea, frozen	1.97	30.0	591.0	174–874
		10.0	197.0	175–875
		3.3	65.0	176–876
Soybean, young	3.28	30.0	984.0	177–877
		10.0	328.0	178–878
		3.3	108.2	179–879
None	....	....	....	180-880
Soybean, medium	2.32	30.0	696.0	181–881
		10.0	232.0	182–882
		3.3	76.6	183–883
Soybean, old	2.15	30.0	645.0	184–884
		10.0	215.0	185–885
		3.3	71.0	186-886
None	....	....	.....	187–887

TABLE 2

Mean probable error and deviations from average checks. Fresh weights on poor soil*

MANURES AFTER BUCKWHEAT		MEAN PERCENTAGE PROBABLE ERROR, SINGLE CROPS										ALL CROPS	
		Buck-wheat	Corn	Corn	Oats	Cotton	Rye	Flax	Millet	Buck-wheat	Corn	Prob-able error	Devia-tion from average check
	gm.												*per cent*
None	....	3.6	3.4	5.2	3.4	5.1	5.6	3.5	8.7	6.7	2.5	4.8	2.1
Pea, medium	30.0	4.0	2.8	5.5	3.4	3.0	4.4	2.0	9.6	5.1	2.4	4.2	46.5
	10.0	5.6	2.7	6.5	4.4	2.9	6.1	2.6	5.6	3.9	2.9	4.3	21.8
	3.3	5.2	3.3	5.4	4.0	3.6	5.1	3.1	7.4	5.4	2.6	4.6	9.6
White Lupin, young	30.0	5.0	4.5	6.0	2.5	5.4	5.3	3.5	9.3	3.9	2.3	4.8	36.2
	10.0	4.5	2.1	5.6	2.9	6.5	3.5	2.8	6.5	5.3	4.4	4.4	14.4
	3.3	3.7	3.2	6.6	3.1	5.2	4.7	2.3	7.4	6.3	3.2	4.6	6.8
None	....	2.7	4.5	6.5	5.2	5.1	6.4	3.1	6.6	5.0	1.9	4.7	−2.0
White Lupin, medium	30.0	2.9	2.2	4.4	4.0	2.7	3.8	4.0	6.8	5.2	2.1	3.8	24.0
	10.0	4.0	3.5	5.1	2.7	3.4	4.0	6.1	11.7	4.3	3.7	4.9	12.5
	3.3	3.3	2.8	5.9	3.1	5.1	4.2	0.6	8.0	4.6	2.2	4.0	6.2
White Lupin, old	30.0	3.0	2.4	2.9	3.6	3.7	3.5	4.5	8.9	4.0	3.5	4.0	23.2
	10.0	3.2	1.2	8.5	6.4	4.5	3.3	0.6	5.6	2.2	2.4	3.8	6.3
	3.3	3.5	2.3	4.8	2.4	4.6	5.7	3.9	6.3	3.0	1.8	3.8	4.9
None	....	3.3	3.2	5.8	3.0	3.8	4.2	4.2	9.7	6.1	2.7	4.6	1.5

Hairy Vetch, young	30.0	5.7	3.4	4.2	3.2	5.0	4.9	3.1	8.3	1.5	3.5	4.3	43.3
	10.0	3.8	1.6	7.5	2.3	4.3	8.4	3.2	4.5	4.1	2.7	4.2	20.6
	3.3	4.6	2.5	6.1	3.4	5.7	6.1	2.9	5.3	4.0	3.0	4.4	11.5
Hairy Vetch, medium	30.0	2.3	1.7	6.8	4.1	5.3	5.6	3.4	9.1	3.9	3.8	4.6	36.8
	10.0	5.2	1.3	5.2	5.2	4.7	4.0	2.9	8.1	2.8	3.2	4.3	16.0
	3.3	3.6	2.2	6.6	2.7	6.2	6.8	3.9	4.8	4.9	2.4	4.4	3.9
Horse Bean, young	10.0	5.2	2.4	4.7	3.2	5.8	5.1	5.3	9.6	3.5	2.6	4.7	13.0
	3.3	3.9	3.9	6.8	1.8	4.0	4.3	3.7	4.9	2.8	2.7	3.9	4.9
None	....	3.0	2.9	4.5	3.1	5.8	5.7	5.7	5.1	5.9	2.9	4.5	−0.8
Horse Bean, old	30.0	3.7	3.6	5.2	1.6	2.8	3.6	3.6	10.1	3.5	1.7	3.9	20.2
	10.0	4.0	2.2	4.3	3.7	4.4	4.6	5.6	5.4	3.9	2.2	4.0	11.5
	3.3	3.8	1.9	8.2	5.5	5.1	3.5	4.2	5.0	5.2	2.9	4.5	1.3
Yellow Lupin, young	30.0	4.3	1.9	7.7	4.2	4.5	4.2	4.2	6.0	2.4	3.2	4.3	32.6
	10.0	3.6	1.8	3.9	2.5	4.3	3.1	2.8	5.7	4.0	2.6	3.4	11.3
	3.3	2.4	2.3	4.7	3.3	6.0	3.6	0.6	6.4	4.4	1.2	3.5	−1.6
None	....	2.8	2.1	8.6	1.9	5.2	2.2	2.9	7.2	4.4	1.9	3.9	−1.4
Yellow Lupin, medium	30.0	5.2	2.0	3.1	1.6	3.9	2.0	2.4	7.3	5.0	3.7	3.6	28.9
	10.0	4.1	2.1	4.0	3.4	4.3	1.5	2.2	7.0	3.9	4.1	3.7	8.9
	3.3	3.6	2.7	5.3	2.3	6.0	3.0	0.6	6.2	4.6	3.1	3.7	2.1
Yellow Lupin, old	30.0	3.3	2.1	3.7	2.5	5.5	4.6	3.5	8.4	2.9	3.6	4.0	2.5
	10.0	4.3	2.5	5.9	2.7	6.0	4.5	0.6	5.1	3.4	3.4	3.3	−0.8
	3.3	2.9	3.9	6.9	2.8	6.6	4.2	4.5	8.0	4.4	1.8	4.6	−5.8
Averages		3.1	3.6	6.1	3.7	4.7	5.0	3.9	7.5	5.6	2.4	4.6	....

* $\left(r = 0.6745 \sqrt{\frac{\Sigma(d)^2}{n\,(n-1)}}\right)$

TABLE 3

Mean probable error and deviation from average check. Fresh weights on rich soil*

MANURES AFTER BUCKWHEAT		MEAN PERCENTAGE PROBABLE ERROR, SINGLE CROPS							ALL CROPS	
		Buckwheat	Corn	Oats	Rye	Millet	Buckwheat	Corn	Probable error	Deviation from average check
	gm.									*per cent*
None	....	6.9	2.1	2.4	1.1	2.1	2.6	1.6	2.7	0.8
Sweet Clover, young	30.0	5.7	2.8	3.6	2.9	3.2	2.2	2.0	3.2	15.3
	10.0	4.4	2.9	3.4	2.7	3.2	2.0	1.8	2.9	7.2
	3.3	5.1	3.5	4.5	3.0	2.8	1.6	1.2	3.1	2.2
Sweet Clover, medium	30.0	3.9	3.1	4.5	3.4	1.5	1.9	1.8	2.9	14.0
	10.0	3.9	2.6	2.2	3.2	2.3	1.6	1.6	2.5	6.5
	3.3	3.7	2.1	2.7	2.2	1.6	1.1	3.2	2.4	3.6
None	....	3.5	3.6	3.4	1.7	2.3	2.6	2.1	2.7	−0.3
Sweet Clover, old	30.0	5.6	2.7	5.8	2.6	2.8	1.5	2.2	3.3	11.2
	10.0	5.4	3.2	4.1	3.8	2.3	2.4	2.0	3.3	6.9
	3.3	3.3	2.6	3.5	3.7	1.5	1.6	2.4	2.7	5.1
Cowpea, young	10.0	4.3	2.7	3.2	3.1	1.4	3.2	2.9	3.0	4.9
	3.3	4.1	3.0	2.8	1.6	2.9	2.4	2.4	2.7	4.2
Cowpea, medium	10.0	3.1	2.8	3.6	3.5	2.9	2.9	1.2	2.9	6.1
	3.3	5.6	1.6	1.8	3.7	1.1	2.1	2.2	2.6	1.9
None	....	4.6	1.7	3.6	1.7	2.6	1.6	1.6	2.5	0.1

Cowpea, old	30.0	5.4	2.9	3.3	3.6	2.2	2.8	3.2	3.3	10.2
	10.0	6.1	3.8	4.4	3.7	1.5	3.1	2.9	3.6	6.2
	3.3	6.1	1.8	4.3	2.9	3.9	2.5	3.0	3.5	4.8
Cowpea, ripe	30.0	3.8	3.7	3.9	3.2	4.8	1.6	1.0	3.1	9.8
	10.0	6.4	2.3	2.1	2.6	3.0	2.4	1.8	2.9	3.5
	3.3	5.4	7.2	3.1	3.8	3.6	2.7	1.9	4.0	−1.3
None	....	6.9	5.0	2.3	2.8	2.6	4.3	3.7	3.9	−0.2
Cowpea, frozen	30.0	7.0	6.8	3.9	2.7	3.1	1.8	3.2	4.1	7.2
	10.0	4.9	7.6	3.4	2.9	3.9	1.5	1.6	3.7	1.3
	3.3	4.0	1.5	2.8	2.4	3.1	2.9	2.5	2.7	3.4
Soybean, young	30.0	4.8	4.2	3.1	2.8	2.7	1.9	3.6	3.3	16.8
	10.0	5.1	4.2	2.7	3.0	2.4	2.0	2.7	3.2	6.2
	3.3	3.9	1.2	2.5	2.6	3.2	3.0	2.8	2.7	5.7
None	....	5.2	6.2	2.9	1.7	2.6	2.5	3.6	3.5	−0.4
Soybean, medium	30.0	4.7	6.2	3.3	3.5	3.4	2.5	2.7	3.8	6.2
	10.0	2.7	2.9	3.8	1.7	3.3	2.5	3.1	2.9	6.1
	3.3	3.4	3.6	2.9	2.1	3.0	3.5	3.7	3.2	4.9
Soybean, old	30.0	4.7	2.7	3.5	2.7	3.0	1.3	1.3	2.7	11.5
	10.0	5.7	4.5	3.4	4.8	4.6	2.9	3.4	4.2	6.1
	3.3	4.6	2.8	2.8	3.9	2.6	2.4	3.2	3.2	2.7
None	....	5.3	2.7	4.5	2.4	1.7	4.5	2.7	3.4	−0.9
Averages	....	5.4	3.6	3.2	1.9	2.3	3.0	2.6	3.1	....

* $\left(r = 0.6745 \sqrt{\frac{\Sigma(d)^2}{n\,(n-1)}}\right)$

probable error calculated for the following crops was the same for the rich and poor soils. The differences shown for the various crops were mainly due to the higher or lower weight of the plants grown.

The final data are always based on the results obtained with sets of 8 pots arranged in rows across the full width of the house. For exact experiments this procedure is of special importance. Frequently vegetation tests in greenhouses are made with duplicate or triplicate sets (7); however, the frequency curves shown in figure 1 for the first buckwheat crop raised on the poor soil demonstrate beyond doubt that accurate results are not to be expected unless the pots receiving the same treatment are evenly distributed over the full width of the house. Even the curves for 6 rows west or 6 rows east show marked deviations from the true average (60 gm.), although 4 of them are identical in both cases.

In the nitrogen availability experiments 1 row of 8 pots was always used for determining the effect of one kind of manuring, but after the check rows, which were scattered throughout

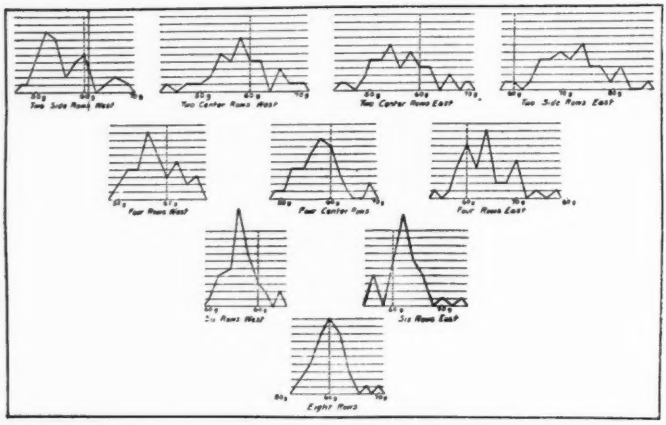

FIG. 1. FLUCTUATIONS OF FRESH WEIGHTS (BUCKWHEAT) HARVESTED IN 2 TO 8 ROWS OF 37 POTS EACH, PLACED LENGTHWISE IN THE HOUSE

Most of the pots in the west-side rows produced smaller crops than those in the east-side rows.

the house, had shown variations in crop yield because of small differences in temperature and ventilation, it seemed preferable to increase the number of parallel tests. Therefore, in subsequent experiments on crop succession, not less than 32 tests were always made in each case, that is, 4 rows of 8 equally treated pots were evenly distributed throughout the house.

Table 4 shows the fluctuations in the fresh weight of the check rows in the first set of experiments. The final figure for each row shows that these slight variations do not materially affect the results obtained with different treatments. Nevertheless, the checking system adopted for the subsequent experiments is undoubtedly preferable, and its use is to be recommended whenever long time tests are planned.

The pots were first watered by daily adjusting their weight on a balance, but it soon became evident that the results did not justify this cumbersome procedure. The desired equality in moisture was no better assured than by careful spraying by well trained men. Occasionally, overwatering will occur, and the total nitrogen balance of the pots of poor

soil proves that in this case nitrogen losses have happened, probably by denitrification in the lower layers of the heavy clay, but again these disturbances by no means impair the

TABLE 4

Deviations in check rows, fresh weights

	AVERAGE CHECK	DEVIATIONS IN ROWS				
		North—Center—South				
North House poor soil						
	gm.					
Buckwheat	60.4	+7.1	−3.9	−6.4	−1.4	+3.6
Corn	122.0	−4.0	−4.0	+7.0	+3.0	−2.0
Corn	24.8	+1.4	+1.2	+2.4	−0.4	−4.4
Oats	62.0	+1.1	−2.8	±0	−2.0	+3.8
Cotton	18.3	−1.7	+0.2	+1.7	+0.7	−1.2
Rye	19.0	+0.1	+2.1	−1.4	−0.4	−0.4
Flax	10.0	−0.3	+0.9	−0.1	−0.9	−0.2
Millet	25.5	+6.5	−1.5	−4.9	+3.7	−3.8
Buckwheat	37.3	+2.9	+1.6	+2.5	−3.0	−4.1
Corn	78.4	−2.0	−4.1	+6.9	−2.1	+1.5
Wheat	8.4	+0.9	+0.1	+0.5	−1.0	−0.4
Total	.....	+18.0	+6.1	+20.5	+7.4	+8.9
	.....	−8.0	−16.3	−12.8	−11.2	−16.5
All crops *gm*	466.1	+10.0	−10.2	+7.7	−3.8	−7.6
All crops *per cent*	100.0	+2.1	−2.2	+1.5	−0.8	−1.4
South House rich soil						
Buckwheat	26.5	+6.2	−1.1	−1.6	−6.4	−2.8	+6.9
Corn	316.0	+2.0	+4.0	−1.0	+2.0	−6.0	−3.0
Oats	111.0	−5.0	−3.0	+3.0	−4.0	+2.0	+3.0
Rye	89.0	−4.0	+4.0	+4.0	+2.0	±0	−9.0
Millet	61.0	−4.0	+2.0	−1.0	+5.0	+1.0	+1.0
Buckwheat	42.2	+1.3	−2.2	+1.3	−0.2	+1.8	−1.7
Corn	62.2	+7.8	−1.2	−3.8	−0.2	+0.8	−2.8
Wheat	42.0	+2.0	±0	.....	.....	±0	−1.0
Total	.....	+19.3	+10.0	+8.3	+9.0	+5.6	+10.9
	.....	−13.0	−7.5	−7.4	−10.8	−8.8	−17.5
All crops *gm*	749.9	+6.3	+2.5	+0.9	−1.8	−3.2	−6.6
All crops *per cent*	100.0	+0.8	+0.3	+0.1	−0.2	−0.4	−0.9

results to such as extent as would be the case with prolonged periods of wet weather in field experiments.

The temperatures in the greenhouse varied to some extent according to the season, as shown in table 5.

During the hottest part of the summer the experiments were usually stopped temporarily,

and the pots were kept dry so that practically no changes occurred in the nitrogen content of the soils.

After the fresh and dry weights of the crops (single pots and rows of 8) had been taken, the total nitrogen for every row of 8 pots was determined by a modified Kjeldahl method with boric acid in the receiving flasks. It proved especially useful for the prompt disposal of the very large number of digestions and distillations that had to be made (27).

TABLE 5

Air temperatures in the greenhouse

	MINIMA			MAXIMA			MEAN TEMPERATURES
	Lowest	Highest	Average	Lowest	Highest	Average	
	°F.	°F.	°F.	°F.	°F.	°F.	°F.
January–March	42	67	56	62	81	71	64
March–May	44	66	57	61	110	84	71
June–July	50	76	65	73	112	96	81
August–September	41	68	60	70	110	89	75
October–December	41	66	53	59	99	77	65

EXPERIMENTAL RESULTS

The plants used as manures in the greenhouse experiments were grown together with a number of other legumes on small field plots at Arlington Farm. Table 6 shows the weights of the air-dried matter and of the total nitrogen calculated in pounds per acre which were harvested in 1915 and 1916.

The data recorded demonstrate clearly that not more than three or four legumes—cowpea, soybean, hairy vetch, and red clover in its second year—have made a really satisfactory growth under the prevailing soil and climate conditions. Field tests continued on the same area for 10 years have confirmed these results, with the exception that red clover has not proved sufficiently reliable. Drought and winter killing have caused about 50 per cent failures. Since only a heavy growth of legumes will benefit the succeeding crops to the fullest extent, such preliminary tests are much to be recommended in view of the widely varying field conditions. No legume can be considered well adapted to a locality if it contains less than 100 pounds nitrogen per acre after full development has been reached.

After the buckwheat test crop was harvested in the greenhouse the pots received different kinds and different quantities of dried and ground legumes as shown in table 1. On poor soil, 10 crops were raised within 4 years, on rich soil, 7 crops within 3 years. The data obtained on fresh weight, dry weight, and nitrogen harvested in the different crops are given in tables 7 to 12. Every figure represents the average of 8 single tests. Percentage calculations on crop increases and nitrogen availability are included.[2]

[2] The figures for the last crop are incomplete, because some of the pots were used for another experiment. The manuring effect had practically reached its end, and the last wheat crop was mainly raised to ascertain this fact conclusively.

TABLE 6

Leguminous crops at different ages

Field tests in 1915 and 1916

	AIR-DRIED SUBSTANCE, POUNDS PER ACRE								NITROGEN, POUNDS PER ACRE							
	Young		Medium		Old		Ripe		Young		Medium		Old		Ripe	
	1915	1916	1915	1916	1915	1916	1915	1916	1915	1916	1915	1916	1915	1916	1915	1916
Alsike	1,590	.....	1,923	1,280	.....	2,051	.....	.....	37.9	....	44.3	38.4	.....	47.2	....	....
Beggarweed	726	770	.....	.....	1,676	1,689	.....	.....	20.5	21.8	.....	.....	28.5	28.7	....	....
Berseem	.....	.....	705	.....	1,410	.....	.....	.....	....	....	20.2	.....	32.4	.....	....	....
Blue Lupin	.....	.....	2,217	.....	.....	.....	.....	.....	....	....	62.3	.....	.....	.....	....	....
Chick pea	.....	.....	1,035	446	1,955	.....	.....	.....	....	....	35.6	11.9	45.4	.....	....	....
Cowpea	768	.....	1,920	5,357	5,397	11,032	3,174	.....	27.2	....	77.6	145.2	140.3	240.5	63.5	....
Crimson Clover	510	.....	2,420	.....	.....	.....	.....	.....	17.1	....	67.3	.....	.....	.....	....	....
Fenngreek	.....	.....	.....	319	.....	.....	.....	938	....	....	.....	11.0	.....	.....	....	21.3
Field Pea	.....	288	991	1,263	.....	1,579	.....	1,269	....	12.1	28.2	44.8	.....	42.3	....	29.6
Hairy Vetch	1,930	2,052	6,017	4,461	.....	4,103	.....	.....	63.1	59.5	163.0	132.0	.....	102.8	....	....
Horse Bean	454	585	.....	.....	1,451	1,964	.....	.....	13.5	22.2	.....	.....	42.2	53.4	....	....
Japan Clover	.....	.....	.....	2,812	.....	.....	.....	.....	....	....	.....	61.3	.....	.....	....	....
Peanut	.....	.....	.....	654	.....	3,461	.....	3,846	....	....	.....	21.7	.....	86.5	....	90.0
Red Clover	.....	1,284	.....	2,799	3,390	4,800	.....	.....	....	30.5	.....	68.2	77.9	129.6	....	....
Sainfoin	.....	.....	.....	640	.....	.....	.....	.....	....	....	.....	11.0	.....	.....	....	....
Seed Vetch	.....	335	.....	1,229	.....	.....	.....	.....	....	12.1	.....	38.5	.....	.....	....	....
Soybean	648	2,400	4,188	5,760	7,778	9,095	5,556	.....	21.3	50.4	82.1	133.6	127.6	195.5	58.9	....
Sweet Clover	624	.....	1,110	2,180	2,560	.....	.....	.....	15.9	.....	29.8	43.2	50.0	.....	....	....
Velvet Bean	.....	2,154	4,800	.....	8,559	.....	2,807	....	....	46.7	115.2	.....	196.0	.....	52.8	....
White Clover	.....	705	.....	855	.....	.....	.....	.....	....	23.2	.....	24.8	.....	.....	....	....
White Lupin	1,110	833	3,856	1,250	2,204	.....	.....	.....	26.3	22.8	68.6	32.8	35.9	.....	....	....
Yellow Lupin	479	.....	1,293	.....	1,116	.....	.....	.....	14.4	....	26.0	.....	19.8	.....	....	....
Yellow Trefoil	.....	.....	876	1,020	.....	.....	.....	.....	....	....	24.4	27.8	.....	.....	....	....

TABLE 7

Fresh weights on poor soil

Averages grams per pot

MANURES		1 CORN	2 CORN	3 OATS	1–3 INCREASE	4 COTTON	5 RYE	6 FLAX	7 MILLET	8 BUCK-WHEAT	9 CORN	4–9 INCREASE	10 WHEAT
	gm.				*per cent*							*per cent*	
None	....	118	26.2	63.1	(−0.2)	16.6	18.1	9.5	33.4	38.0	76.4	(+3.0)	9.3
Pea, medium	30.0	233	47.5	97.4	85.1	19.3	24.3	11.1	44.9	42.5	88.0	25.3	10.9
	10.0	188	33.4	74.3	44.0	18.2	21.5	9.2	31.0	35.5	81.6	7.0	....
	3.3	151	23.2	69.3	17.9	16.1	19.4	9.1	28.0	39.0	79.6	4.0	....
White Lupin, young	30.0	201	50.6	90.1	67.0	18.2	24.6	10.4	32.0	40.4	88.3	16.3	10.8
	10.0	152	31.8	75.0	25.6	18.5	19.6	10.0	31.9	34.6	80.5	6.0	....
	3.3	128	32.8	70.4	11.8	19.8	20.4	9.2	27.0	37.3	78.9	5.1	....
None	....	118	26.0	59.2	(−0.2)	18.5	20.1	10.1	25.4	36.7	74.3	(+0.5)	8.5
White Lupin, medium	30.0	183	38.5	77.2	45.5	20.0	21.0	10.6	26.1	38.5	84.4	14.5	9.4
	10.0	147	31.4	76.4	23.6	20.1	18.3	10.3	23.8	36.0	78.3	1.4	....
	3.3	136	26.5	68.6	11.7	19.8	16.6	9.7	24.4	36.0	81.0	1.8	....
White Lupin, old	30.0	175	38.7	78.4	41.1	21.2	19.6	10.6	26.2	40.8	86.6	11.5	9.6
	10.0	135	24.4	74.2	12.9	20.0	17.0	10.2	25.8	39.0	84.9	7.0	....
	3.3	129	26.6	68.1	8.0	18.4	17.7	9.6	27.1	38.5	82.3	5.1	....
None	....	129	27.2	62.0	(+5.2)	20.3	16.6	9.1	22.0	37.6	85.3	(−1.2)	8.9
Hairy Vetch, young	30.0	234	57.2	88.8	86.2	21.8	20.4	11.2	27.1	42.8	86.3	13.5	9.1
	10.0	185	31.8	74.4	41.8	19.4	19.9	9.9	23.2	43.6	82.6	8.0	....
	3.3	151	24.9	74.5	21.4	20.1	17.0	9.5	20.1	41.1	84.9	4.7	....

Hairy Vetch, medium	30.0	227	43.9	81.2	72.2	22.5	19.7	11.5	30.1	42.8	91.3	18.5	9.1
	10.0	157	31.0	75.7	28.0	21.6	19.9	10.0	24.4	42.1	86.1	11.2	8.5
	3.3	135	24.6	62.0	7.0	21.0	17.7	9.9	23.1	36.6	83.0	4.0	8.1
Horse Bean, young	10.0	158	31.3	72.0	26.8	19.8	18.2	10.1	25.6	38.1	81.5	5.0	8.0
	3.3	137	26.6	64.0	10.0	18.8	16.6	9.6	26.6	35.5	81.3	2.4	8.0
None	....	125	24.4	60.0	(+0.8)	19.0	17.6	8.3	30.6	32.1	76.3	(−0.1)	7.4
Horse Bean, old	30.0	172	38.9	80.6	41.9	20.4	19.9	10.4	29.3	38.1	80.9	8.2	8.0
	10.0	153	30.4	69.4	27.5	18.6	18.1	9.8	25.6	33.6	84.0	3.0	7.5
	3.3	133	21.6	67.2	7.0	18.4	17.7	9.1	26.9	33.6	74.4	−2.2	7.6
Yellow Lupin, young	30.0	214	39.8	87.0	66.6	18.9	20.0	9.9	24.4	35.8	96.6	17.5	9.0
	10.0	163	26.9	68.3	25.3	17.7	18.2	10.3	28.5	34.8	81.5	3.8	7.9
	3.3	127	21.8	59.6	0.4	17.1	17.3	9.5	22.4	35.5	78.1	−2.3	7.5
None	....	120	20.4	65.8	(−0.7)	17.1	17.6	9.0	23.1	31.0	79.9	(−3.5)	8.0
Yellow Lupin, medium	30.0	186	40.0	88.0	53.2	19.9	23.8	10.1	28.0	37.4	90.5	13.0	8.9
	10.0	142	28.1	65.4	13.9	19.7	21.2	10.1	26.8	35.1	82.5	0.8	7.9
	3.3	126	24.0	61.9	2.1	22.1	18.3	10.1	26.0	35.3	81.4	5.0	7.9
Yellow Lupin, old	30.0	110	31.2	76.0	4.7	21.2	21.2	9.7	26.7	34.4	79.5	4.7	8.6
	10.0	109	24.1	69.8	−2.4	21.5	19.0	10.0	30.8	36.4	73.6	3.9	8.5
	3.3	104	21.8	66.6	−7.1	19.5	19.0	9.9	26.8	34.4	70.8	−2.0	9.0
Average checks		122	24.8	62.0	....	18.3	18.0	9.2	26.9	35.1	78.4	....	8.4

TABLE 8

Fresh weights on rich soil

Averages grams per pot

MANURES		1 CORN	1 INCREASE	2 OATS	3 RYE	4 MILLET	5 BUCK-WHEAT	6 CORN	2-6 INCREASE	7 WHEAT
	gm.		per cent						per cent	
None	....	318	(+0.7)	106	85	57	43.5	70	(−1.2)	44
Sweet Clover, young	30.0	396	26.7	108	99	66	43.5	67	5.8	44
	10.0	372	18.7	102	91	59	43.0	68	−2.1	..
	3.3	334	6.0	109	88	61	42.0	67	0.4	..
Sweet Clover, medium	30.0	403	29.0	110	99	67	42.5	65	5.0	..
	10.0	368	14.0	98	95	66	42.0	64	0.1	36
	3.3	339	7.7	106	95	62	44.5	64	1.7	39
None	....	320	(+1.3)	108	93	63	40.0	61	(±0)	42
Sweet Clover, old	30.0	385	23.0	115	100	62	44.5	61	4.8	..
	10.0	359	14.3	112	96	64	42.5	63	3.6	..
	3.3	346	10.0	108	101	62	42.0	63	2.9	..
Cowpea, young	10.0	366	16.7	103	103	62	43.5	64	2.7	44
	3.3	349	11.0	106	100	60	44.5	61	1.7	41
Cowpea, medium	10.0	353	12.3	112	94	63	46.5	65	4.2	45
	3.3	347	10.3	110	94	65	43.5	61	2.2	..
None	....	315	(−0.3)	144	93	60	43.5	58	(+1.1)	..

Cowpea, old	30.0	366	16.7	112	110	71	44.5	61	9.2	..
	10.0	359	14.3	110	94	71	44.0	61	4.1	39
	3.3	337	7.0	112	97	65	45.5	56	2.7	41
Cowpea, ripe	30.0	356	13.3	125	101	66	48.0	67	11.6	45
	10.0	338	7.7	112	94	65	44.5	65	4.2	..
	3.3	305	−3.7	120	91	65	45.0	62	4.9	..
None	....	318	(+0.7)	107	91	66	42.0	62	(−0.4)	..
Cowpea, frozen	30.0	331	5.0	113	99	73	43.5	60	6.4	41
	10.0	328	4.0	114	87	68	43.0	65	3.2	41
	3.3	305	−3.7	112	91	64	45.0	66	3.5	41
Soybean, young	30.0	418	34.0	112	105	73	44.0	63	8.7	..
	10.0	374	19.3	106	91	64	41.5	65	0.6	..
	3.3	362	15.3	113	89	65	43.5	63	2.2	..
None	....	310	(−2.0)	113	89	62	44.0	63	(+1.6)	42
Soybean, medium	30.0	380	21.3	116	99	68	44.5	68	8.3	44
	10.0	359	14.3	105	100	65	42.5	64	3.1	43
	3.3	347	10.3	109	103	62	42.0	64	4.0	..
Soybean, old	30.0	378	17.3	112	95	73	44.0	69	7.6	..
	10.0	352	12.0	111	88	70	44.5	63	3.1	..
	3.3	343	9.0	104	86	69	43.5	65	0.6	44
None	....	313	(−1.0)	116	80	62	40.5	59	(−2.2)	41
Average checks		316	....	111	89	61	42.2	62.2	....	42

TABLE 9

Dry weights on poor soil

Averages grams per pot

MANURES		1 CORN	2 CORN	3 OATS	1–3 INCREASE	4 COTTON	5 RYE	6 FLAX	7 MILLET	8 BUCK-WHEAT	9 CORN	4–9 INCREASE	10 WHEAT
	gm.				*per cent*							*per cent*	
None	....	19.2	4.0	15.4	(−0.8)	3.8	5.1	1.9	16.1	5.9	21.6	(+1.1)	2.2
Pea, medium	30.0	38.6	6.7	23.0	76.4	5.3	6.7	2.3	20.8	7.0	25.5	7.2	2.5
	10.0	33.2	4.4	18.0	43.4	4.8	6.3	2.1	14.4	5.1	23.5	1.0	...
	3.3	26.4	3.3	16.8	20.1	4.3	5.3	1.9	12.8	5.4	23.3	0.4	...
White Lupin, young	30.0	33.5	7.3	21.2	60.1	4.3	7.5	2.1	15.0	5.8	25.4	3.9	2.4
	10.0	24.8	4.4	18.2	27.3	4.6	5.8	2.0	16.0	5.1	23.4	2.5	...
	3.3	20.7	4.9	17.4	10.7	4.8	6.0	1.9	11.8	5.7	23.0	0	...
None	....	18.9	3.8	15.1	(−2.9)	4.7	5.0	2.1	11.5	5.4	22.5	(+0.5)	1.9
White Lupin, medium	30.0	29.8	5.7	18.8	40.0	5.5	6.1	2.1	11.1	5.7	23.8	1.0	2.2
	10.0	25.4	4.7	19.2	27.0	5.3	5.4	2.0	9.1	4.9	21.8	−1.8	...
	3.3	22.7	4.0	17.0	12.5	4.9	5.3	2.0	12.6	4.7	23.4	0.4	...
White Lupin, old	30.0	29.1	5.4	19.2	38.5	5.7	5.8	2.1	11.5	6.1	24.9	1.9	2.1
	10.0	21.4	3.9	17.9	11.2	5.0	5.1	2.0	11.9	5.5	24.5	−1.6	...
	3.3	20.0	3.9	16.3	3.4	5.0	5.2	1.9	11.5	5.6	24.0	0.7	...
None	....	19.4	3.9	15.0	(−1.6)	5.1	4.9	1.8	10.4	5.9	24.0	(+0.1)	2.1
Hairy Vetch, young	30.0	35.5	9.2	21.9	72.0	5.7	6.6	2.3	12.0	6.6	25.1	3.0	2.4
	10.0	33.2	4.4	17.6	42.4	4.9	5.8	2.1	10.4	6.6	23.5	0.6	...
	3.3	23.3	3.6	18.2	3.1	5.1	5.0	2.0	9.0	6.6	25.1	0.3	...

Hairy Vetch, medium	30.0	35.7	6.8	19.9	61.1	6.0	5.8	2.2	14.9	7.1	26.0	4.9	2.2
	10.0	26.9	4.4	18.3	30.4	5.5	6.1	2.2	10.8	6.8	25.0	2.1	2.0
	3.3	20.5	3.8	14.7	0.3	5.2	5.4	2.1	9.9	6.0	24.9	0.6	1.8
Horse Bean, young	10.0	26.2	4.6	17.2	23.7	5.3	5.2	2.0	11.3	5.3	24.3	0.6	1.9
	3.3	22.6	3.8	15.3	7.3	5.1	5.1	2.0	12.4	5.2	23.6	0.6	1.8
None	....	22.2	2.9	15.0	(+0.5)	4.9	5.1	1.7	15.0	4.8	21.1	(+0.2)	1.8
Horse Bean, old	30.0	29.8	5.7	19.5	41.9	5.5	5.6	2.2	13.5	5.7	24.5	2.4	1.9
	10.0	26.2	4.1	16.7	23.7	5.1	5.2	2.0	12.9	4.8	25.0	1.4	1.9
	3.3	19.6	2.9	16.3	−0.3	4.8	5.2	1.9	12.3	5.1	22.1	−0.4	1.9
Yellow Lupin, young	30.0	35.5	5.7	21.2	61.1	5.2	5.8	2.1	10.8	5.6	26.8	2.0	2.2
	10.0	30.5	3.8	16.5	10.9	4.8	5.4	2.1	13.8	5.1	25.6	2.3	1.9
	3.3	21.2	3.3	14.3	−0.3	4.8	5.1	2.0	8.3	5.4	23.1	−1.7	1.9
None	....	20.9	2.9	15.8	(+1.8)	4.3	5.0	1.8	11.1	5.2	23.4	(−0.7)	1.9
Yellow Lupin, medium	30.0	33.2	5.1	21.4	55.7	5.5	6.4	2.2	13.0	5.4	26.8	3.5	1.9
	10.0	25.9	4.0	15.9	43.9	5.1	5.6	2.1	12.1	5.0	23.9	0.8	1.9
	3.3	20.8	3.7	15.5	2.9	5.5	4.6	2.1	12.1	5.0	23.4	0.2	1.9
Yellow Lupin, old	30.0	18.2	4.4	21.3	13.0	5.2	5.7	2.0	13.3	5.3	23.6	1.4	2.0
	10.0	18.9	3.4	20.2	9.4	5.2	5.2	2.2	13.6	5.3	22.4	0.8	2.0
	3.3	17.9	3.1	18.9	2.6	4.2	4.6	2.0	13.3	5.6	20.4	−0.5	2.2
Average checks		20.1	3.5	15.3	....	4.6	5.0	1.8	12.8	5.5	22.5	....	2.0

TABLE 10

Dry weights on rich soil

Averages grams per pot

MANURES		1 CORN	1 INCREASE	2 OATS	3 RYE	4 MILLET	5 BUCK-WHEAT	6 CORN	2–6 INCREASE	7 WHEAT
	gm.		*per cent*						*per cent*	
None	....	88.3	(+9.2)	38	29	20	4.8	24.6	(±0)	11.9
Sweet Clover, young	30.0	117.8	46.1	41	34	22	5.0	21.6	6.2	11.9
	10.0	106.1	31.5	37	31	20	5.3	22.5	4.6	....
	3.3	98.3	21.7	40	30	21	5.0	22.8	2.0	....
Sweet Clover, medium	30.0	108.3	34.2	40	34	23	5.1	21.3	6.1	....
	10.0	102.0	26.5	38	33	22	4.9	20.3	3.3	10.9
	3.3	93.3	15.5	39	33	21	5.1	21.3	2.6	10.9
None	....	80.5	(−0.5)	39	32	22	4.6	19.3	(+0.3)	11.5
Sweet Clover, old	30.0	107.5	33.3	43	35	21	5.1	19.3	6.1	....
	10.0	95.5	18.2	40	33	21	5.3	19.3	1.7	....
	3.3	91.3	13.0	39	35	21	5.3	19.0	2.4	....
Cowpea, young	10.0	104.3	29.2	37	35	21	5.5	18.8	0.7	12.0
	3.3	92.8	14.9	37	34	20	5.9	17.4	−2.0	11.9
Cowpea, medium	10.0	101.1	25.2	40	32	21	6.1	18.6	1.0	12.5
	3.3	90.5	12.0	40	33	22	5.6	17.6	1.5	....
None	....	79.6	(−1.6)	41	32	20	5.6	17.4	(+0.4)	....

Cowpea, old	30.0	105.6	30.9	40	37	24	5.5	18.0	6.3	....
	10.0	96.3	19.2	40	32	24	5.4	17.5	2.1	11.6
	3.3	88.5	9.5	39	33	22	6.0	15.3	1.0	11.5
Cowpea, ripe	30.0	101.4	25.6	45	35	22	6.1	20.6	10.7	12.4
	10.0	93.3	15.5	39	32	22	5.6	18.1	0.3	....
	3.3	88.4	9.4	40	31	22	6.3	17.1	0	....
None	....	72.1	(−11.0)	39	30	22	5.1	17.5	(+2.5)	....
Cowpea, frozen	30.0	102.2	26.6	38	34	26	5.6	17.8	4.3	12.0
	10.0	95.1	17.7	42	30	24	5.4	18.9	3.4	12.1
	3.3	90.2	11.7	40	31	23	6.0	18.9	2.1	11.0
Soybean, young	30.0	98.7	22.3	41	37	25	5.5	17.8	8.1	....
	10.0	96.7	19.7	38	31	22	5.4	20.1	0	....
	3.3	91.1	12.7	41	31	22	5.8	17.0	0.3	....
None	....	82.8	(+2.4)	40	31	23	5.6	19.6	(+2.4)	12.1
Soybean, medium	30.0	103.7	28.5	42	34	23	5.9	20.1	7.4	12.5
	10.0	94.6	17.1	37	35	23	5.5	18.0	1.7	12.6
	3.3	88.0	8.9	39	36	21	5.1	18.8	3.0	....
Soybean, old	30.0	100.1	24.0	41	32	26	5.6	19.4	6.6	....
	10.0	94.3	16.7	39	31	25	5.4	19.3	2.8	....
	3.3	88.7	9.7	36	30	24	5.5	20.3	−0.6	13.0
None	....	79.4	(−1.9)	41	28	22	5.3	17.1	−1.0	11.6
Average checks		80.9	....	40	30	22	5.2	19.3	...	11.8

TABLE 11

Nitrogen in crops on poor soil

Averages milligrams per pot

NITROGEN APPLIED		1 CORN	2 CORN	3 OATS	1–3 RETURN	4 COTTON	5 RYE	6 FLAX	7 MILLET	8 BUCK-WHEAT	9 CORN	10 WHEAT	1–10 RETURN
	mgm.				*per cent*								*per cent*
None	.....	113.3	45.2	98.6	....	60.4	80.3	38.6	66.7	59.0	99.4	31.2	....
Pea, medium	855.0	359.0	71.7	138.0	33.3	100.7	110.9	41.4	97.5	64.4	104.6	35.5	47.0
	285.0	212.5	51.0	117.0	36.5	82.6	91.1	37.1	82.7	48.5	103.4	....	53.6
	94.1	153.6	40.3	102.5	32.7	83.6	78.3	33.3	70.4	51.8	100.2	....	51.9
White Lupin, young	711.0	251.2	82.5	152.6	30.1	88.6	117.8	39.1	70.5	56.2	104.1	38.1	41.4
	237.0	161.2	52.4	123.8	28.7	96.1	84.7	37.4	78.4	52.0	100.6	....	49.5
	78.2	134.5	51.9	116.6	45.5	85.9	89.1	31.0	61.4	55.9	94.3	....	71.9
None	.....	134.2	39.1	93.6	....	83.4	78.8	34.8	51.7	58.9	92.3	30.2	....
White Lupin, medium	534.0	196.7	67.3	135.4	23.8	105.6	84.8	37.2	73.3	66.1	97.6	36.1	35.8
	178.0	160.0	57.3	130.6	37.4	108.1	76.7	34.4	54.5	54.9	91.6	....	54.1
	58.7	144.6	50.5	110.5	47.9	89.7	72.8	35.0	63.0	46.5	88.9	....	71.4
White Lupin, old	489.0	189.1	67.4	136.3	24.4	112.9	83.5	35.1	58.7	67.1	94.6	32.8	34.7
	163.0	137.0	49.6	127.1	33.3	80.5	72.4	32.2	60.7	60.5	93.1	....	38.1
	53.8	118.0	47.6	115.7	52.3	82.0	72.8	36.9	66.1	54.3	88.8	....	71.9
None	.....	122.2	45.2	106.5	....	100.0	69.5	31.5	52.0	65.5	98.4	33.0	....
Hairy Vetch, young	981.0	404.7	106.2	164.3	42.9	114.6	93.7	41.4	60.0	82.5	107.9	36.0	53.6
	327.0	255.6	62.4	128.5	56.4	102.4	77.1	35.5	51.0	69.3	103.4	....	68.4
	107.9	151.5	53.4	123.8	62.4	105.1	86.0	35.3	50.5	66.0	95.4	....	89.7

Hairy Vetch, medium	813.0	382.0	72.8	139.3	40.8	124.2	83.5	48.2	46.5	69.6	119.6	31.7	63.1
	271.0	191.0	49.3	146.4	45.9	109.5	82.4	41.8	63.5	72.1	110.0	28.0	72.3
	89.4	145.5	42.9	102.9	30.8	105.0	74.9	38.2	55.5	60.6	104.6	27.2	69.7
Horse Bean, young	297.0	188.7	52.9	120.4	34.1	121.9	74.9	38.8	64.4	62.0	104.5	28.1	53.8
	98.0	180.8	41.0	93.3	46.4	89.8	65.3	36.5	61.4	52.5	96.8	28.0	57.1
None	....	139.8	32.5	91.5	....	90.2	71.9	29.6	65.0	52.8	99.2	28.0	....
Horse Bean, old	873.0	232.5	66.1	128.7	26.7	104.0	78.1	44.2	75.1	64.4	115.2	30.0	35.1
	291.0	175.5	47.2	116.9	26.2	102.0	72.3	36.8	74.4	54.2	105.0	30.0	41.4
	96.0	125.4	36.2	110.8	9.9	92.0	70.7	37.4	66.4	52.0	92.8	29.0	32.4
Yellow Lupin, young	900.0	312.4	69.0	148.4	29.8	90.0	82.4	38.4	68.4	59.9	120.6	33.2	33.9
	300.0	192.1	44.8	107.3	25.4	92.6	73.4	37.0	57.2	58.1	107.5	29.8	33.0
	99.0	123.0	40.0	94.4	0.5	92.6	72.5	44.9	41.5	60.5	108.6	29.8	33.0
None	.....	133.8	33.4	90.1	....	80.0	70.7	29.9	61.1	63.4	107.6	29.5	....
Yellow Lupin, medium	603.0	242.4	61.2	145.5	29.0	106.7	97.3	40.5	64.2	62.1	104.5	29.8	40.2
	201.0	168.4	46.4	106.5	25.8	101.5	89.0	48.9	61.2	53.5	105.2	28.9	54.6
	66.3	118.6	37.4	113.2	32.0	109.5	73.9	37.6	67.0	53.5	103.0	27.6	73.1
Yellow Lupin, old	381.0	121.9	50.6	153.4	15.1	110.8	86.6	40.6	52.0	63.1	94.4	30.0	29.7
	127.0	117.2	40.8	133.3	19.0	97.8	82.2	40.5	75.6	53.5	89.6	29.8	43.6
	41.9	100.2	40.9	134.2	9.7	89.9	72.0	37.8	72.8	57.1	89.8	33.4	50.0
Average checks		128.7	39.1	96.1	....	82.2	74.2	32.9	59.3	59.9	99.4	30.5	....

TABLE 12

Nitrogen in crops on rich soil

Averages milligrams per pot

NITROGEN APPLIED		1 CORN	RETURN	2 OATS	3 RYE	4 MILLET	5 BUCK-WHEAT	6 CORN	7 WHEAT	1–7 RETURN
	mgm.		*per cent*							*per cent*
None	.....	593.5	.....	209.0	203.0	106.0	86.3	206.7	178.5	.....
Sweet Clover, young	762.0	1017.3	55.0	291.1	244.7	125.4	97.5	187.9	178.5	75.2
	254.0	773.7	66.2	218.3	229.3	126.0	97.6	209.1	.....	87.0
	83.8	605.7	36.4	220.0	201.0	123.9	93.0	203.0	.....	79.3
Sweet Clover, medium	804.0	876.2	34.5	208.0	227.9	151.1	101.6	200.2	.....	41.9
	268.0	719.9	55.8	207.0	231.0	129.8	93.0	186.6	176.6	79.2
	88.4	633.6	66.0	171.6	234.3	119.8	95.5	204.5	167.9	103.7
None	.....	586.3	.....	210.6	217.7	129.8	86.8	206.3	119.6	.....
Sweet Clover, old	693.0	737.0	21.9	245.1	248.5	123.9	91.8	216.2	.....	38.8
	213.0	618.0	22.1	204.0	221.0	113.4	104.5	179.2	.....	27.7
	70.3	624.0	38.8	226.2	231.0	111.4	101.3	182.3	.....	89.0
Cowpea, young	354.0	700.1	32.2	192.4	235.0	117.6	113.2	172.7	134.4	43.6
	116.8	635.6	53.1	222.0	241.0	104.0	108.5	198.4	133.3	102.5
Cowpea, medium	271.0	597.1	8.1	224.0	230.0	126.0	114.6	180.2	140.0	27.1
	89.4	562.3	2.9	192.0	228.0	132.0	103.5	193.6	.....	41.6
None	.....	575.9	.....	237.8	218.0	120.0	103.5	184.3	.....	.....

Cowpea, old	654.0	755.8	27.5	180.0	255.0	132.0	110.5	190.9	.....	38.1
	218.0	692.6	54.4	196.0	214.0	127.2	98.3	185.4	116.0	60.8
	71.9	597.7	51.9	206.7	247.0	118.8	118.8	174.4	119.0	127.7
Cowpea, ripe	600.0	709.9	24.8	207.7	252.0	138.6	111.6	214.1	136.4	42.3
	200.0	668.9	46.9	206.7	230.0	121.0	108.5	190.1	.....	70.1
	66.0	645.0	117.4	200.0	226.0	123.2	115.9	184.7	.....	171.6
None	.....	576.2	.....	179.4	216.0	103.4	92.8	183.8	.....	.....
Cowpea, frozen	591.0	775.0	35.2	186.2	228.0	145.4	101.8	199.3	112.8	44.0
	197.0	691.0	64.5	222.6	228.0	127.2	97.2	200.2	96.8	87.5
	65.0	697.5	149.4	192.0	217.0	126.5	121.8	194.8	124.3	206.9
Soybean, young	984.0	1178.8	62.1	172.2	259.0	109.9	108.3	185.1	.....	67.5
	328.0	876.8	96.4	205.2	223.0	121.0	104.8	194.9	.....	101.8
	108.2	677.4	114.5	207.7	223.0	121.0	108.4	192.1	.....	136.9
None	.....	567.7	.....	188.0	208.0	119.6	109.7	199.9	118.6	.....
Soybean, medium	696.0	785.6	88.9	277.2	228.0	138.0	103.7	192.9	150.0	45.1
	232.0	673.4	40.0	173.9	245.0	151.8	112.1	181.8	153.7	78.4
	76.6	670.3	102.8	187.1	252.0	100.9	100.6	180.4	.....	157.5
Soybean, old	645.0	730.0	25.4	217.3	221.0	111.8	113.6	221.1	.....	33.2
	215.0	646.8	27.7	179.2	217.0	122.4	105.3	216.1	.....	46.6
	71.0	636.5	79.4	215.0	207.0	103.2	112.6	196.8	184.0	181.2
None	.....	601.7	.....	221.4	196.0	110.0	106.0	174.3	148.5	.....
Average checks		575.2	.....	207.7	216.7	114.8	97.5	192.6	141.3	.....

The percentage deviation of the total fresh weight of all manured crops from the average check is given in tables 2 and 3, together with the mean probable error calculated for all crops. A comparison of these figures with one another shows that the crop increases caused by the smallest application of green manures (equivalent to about 25 pounds nitrogen per acre) are in most cases not much larger, sometimes even smaller, than the single probable error. Undoubtedly the accuracy of these results is not very high, as is to be expected when such small quantities of organic nitrogen are applied.

Tables 7 to 12 show that the original productivity of the rich soil was about twice as high as that of the poor soil and that the total average production of all crops (including the buckwheat used as preliminary test crop) was on the unmanured check rows: on poor soil, 99.6 gm. dry weight with 810.8 mgm. N per pot; on rich soil, 211.9 gm. dry weight with 1638.1 mgm. N per pot.

TABLE 13

Average crop increases and nitrogen return on poor and on rich soil

MANURE PER POT	DRY WEIGHTS, PER CENT INCREASE				NITROGEN, PER CENT RECOVERED			
	Poor soil		Rich soil		Poor soil		Rich soil	
	Crops 1–3	Crops 4–9	Crop 1	Crops 2–6	Crops 1–3	Crops 1–10	Crop 1	Crops 1–7
gm.								
30.0	51.0	3.1	30.2	6.9	29.4	41.5	35.0	47.5
10.0	26.7	1.1	21.5	2.0	33.5	54.8	46.8	64.5
3.3	5.7	0.3	12.6	1.4	33.6	61.2	73.9	127.1
Averages...	27.8	1.5	21.4	3.4	32.2	52.5	51.9	79.7

Accordingly, the crop increases caused by green manuring were larger and more lasting on the poor soil, especially with the heaviest application of legumes. The average percentage increase in dry weights and the average nitrogen return calculated from the data in tables 9 to 12 are shown in table 13.

On poor soil the first 3 crops were distinctly benefited by the green manuring, whereas very little effect was noticeable with the following 6 crops. On rich soil the first crop only was markedly influenced, and after 5 other crops were taken the effect had practically reached its end. The relations selected for the different quantities of green manures (9:3:1) are very clearly reflected in the crop increases on poor soil. Those for the total fresh weights are 7.5:3:1 and the relations of the dry weight increases in the fourth to ninth crop (3.1:1.1:0.3) are almost identical with those chosen for the green manures (30:10:3.3). On the rich soil the crop increases are disproportionately large with the smaller applications of green manures, obviously because of an enhanced mobilization of soil nitrogen, caused by more vigorous bacterial action in the manured pots.

The detailed figures contained in tables 11 and 12 show that with large applications of green manures the nitrogen returns are fairly similar with different plants and on different soils. With smaller applications, however, the rich

TABLE 14

Nitrification and nitrogen availability of green manures in poor and in rich soil

GREEN MANURES		N NITRIFIED	N RECOVERED PER CROP
Poor soil			
		per cent	*per cent*
Pea, medium		22	25
White Lupin	Young	8	10
	Medium	14	14
	Old	7	6
Hairy Vetch	Young	29	31
	Medium	25	28
Horse Bean	Young	15	32
	Old	19	12
Yellow Lupin	Young	8	13
	Medium	5	13
	Old	6	..
Averages		14	16
Rich soil			
Sweet Clover	Young	19	52
	Medium	20	52
	Old	12	28
Cowpea	Young	33	28
	Medium	19	2
	Old	8	45
	Ripe	5	65
	Frozen	3	80
Soybean	Young	30	91
	Medium	23	57
	Old	16	44
Averages		17	50

soil is far superior; the nitrogen return rises in one case to more than 200 per cent. That the green manure nitrogen is more completely mineralized by the soil organisms, and that it is of comparatively greater effect upon the manured

plants when it is applied in moderate quantities, is indicated by the average percentage of nitrogen recovered on both soils after large, medium, and small applications, viz., 42. 55, and 61 per cent on the poor soil, 47, 65, and 127 per cent on the rich soil.

The results of nitrification tests presented in table 14 furnish additional information concerning the different behavior of the green manures in poor and in rich soil.

In poor soil, percentage nitrification and nitrogen return in the first crop agree very closely in most cases. Although in the nitrification tests approximately three times the maximum amount of green manure pots was used, the results obtained for the poor soil confirm the frequently recorded fact that nitrification and vegetation experiments may give similar results despite the different conditions under which they are made. With the rich soil, on the other hand, the nitrification tests gave results far below the nitrogen returns in the first crop. A comparison of the nitrification results for both soils, however, shows an almost identical behavior of the different materials, and a marked reduction in nitrate formation when more mature legumes were tested. The conclusion is justified, that the actual availability of green manure nitrogen was more accurately measured in the first crop by the nitrification experiments than by the vegetation tests.

Additional information on the increase in the humus decomposition in the rich soil, apparently caused by the accelerated microbial action in the soil lightly dressed with green manure, was secured from a special set of nitrification experiments in which the quantities of green manure applied agreed closely with those used in the pot tests; 0.3, 0.1, and 0.03 gm. dry substance were added to 100 gm. soil, respectively, with 12, 4, and 1.2 mgm. total nitrogen. Dried young vetch, an easily nitrifiable material, was added to the soil, and 6 tests were made in every case. The following confirmative results were obtained (per cent nitrogen nitrified): large quantity, 50.4; medium quantity; 68.3; small quantity, 195.1.

The stimulation of humus decomposition in the manured soils is shown likewise by the average nitrogen returns recorded graphically for the successive crops. (Fig. 2.) The main parts of the resulting curves reflect very clearly the gradually diminishing returns, but the initial deviations from these curves shown by the first crops leave no doubt that some other source of nitrogen must have caused this irregularity. That the humus nitrogen is responsible for this effect is further evidenced by the different behavior of the two soils. In contrast to the poor soil, the rich soil, because of its higher nitrogen content, shows no deviation from the curve to the negative side. The initial increase in the mineralization of soil nitrogen and its utilization by the first crop is followed by a compensating deviation from the curve to the negative side, indicating a temporary exhaustion of available soil nitrogen.

The gradual flattening of the availability curve in the third and fourth year after the green manures had been applied demonstrates very convincingly that in soils not exceptionally rich in humus nitrogen a 50 to 60 per cent

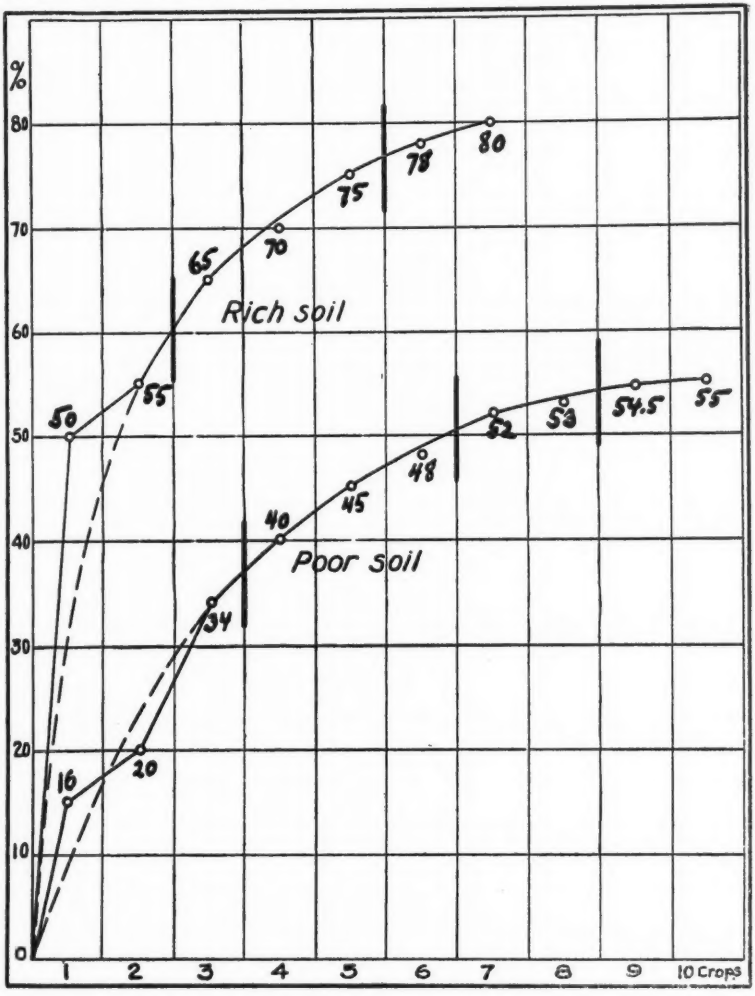

FIG. 2. AVERAGE NITROGEN RETURNS (PER CENT) IN 10 CROPS ON POOR SOIL AND IN 7 CROPS ON RICH SOIL

The short vertical lines separate the years in which the different crops were grown

nitrogen availability can be considered as an average return for green manuring. Soils poor in humus nitrogen naturally induce humus formation, and in them part of the organic nitrogen is firmly maintained in this form by microbial

TABLE 15

Percentage of nitrogen recovered in successive crops

GREEN MANURE DRY	CROP NUMBER	POOR SOIL												RICH SOIL										
		Pea	Hairy Vetch		Horse Bean		White Lupin			Yellow Lupin			Sweet Clover			Cowpea					Soybeans		
		Medium	Young	Medium	Young	Old	Young	Medium	Old	Young	Medium	Old	Young	Medium	Old	Young	Medium	Old	Ripe	Frozen	Young	Medium	Old
5%	1	27	29	31	..	20	16	11	10	19	17	..	55	35	22	...	...	28	25	35	62	29	25
	2	4	7	4	..	3	6	5	6	3	4	..	11	...	6	...	..	...	...	...	...	10	...
	3	5	7	5	..	..	8	7	8	6	8	15	4	1	5	...	..	6	6	2	4	2	1
	4	2	3	5	..	4	1	4	6	1	4	8	1	4	2	...	..	3	4	5	...	3	...
	5	4	2	11	..	3	6	2	2	1	4	3	..	1	..	...	..	2	4	1	1	1	3
30	6	..	1	2	..	..	1	1	..	1	1	2	..	1	4	...	..	...	4	1	...	...	4
	7	5	..	1	..	2	2	3	..	1	1	..	4	...	..	...	..	...	...	...	...	...	...
	8	..	3	..	..	2	..	1	2	..	..	1	..	...	..	...	..	...	...	...	...	...	...
	9	..	1	3	..	2	..	..	..	2	1	..	..	...	..	...	..	...	...	...	...	...	...
	10	..	..	..	..	..	1	1	1	..	..	..	..	...	..	...	..	...	...	...	...	...	...
	Total...	47	53	62	..	36	41	35	35	34	40	29	75	42	39	...	..	39	43	44	67	45	33
	1	25	40	24	21	16	11	8	7	20	17	..	66	56	22	32	4	54	54	65	96	40	28
	2	4	7	4	5	3	6	10	6	2	3	..	4	...	..	...	6	...	1	7	...	...	...
	3	7	10	18	8	7	12	19	18	4	5	19	5	6	2	6	5	...	7	6	2	12	1
	4	..	6	10	13	7	6	14	..	3	10	12	5	6	..	1	5	6	3	6	1	16	4
	5	6	1	3	..	..	4	2	1	..	7	6	..	...	4	5	7	...	6	...	2	7	4
10	6	2	..	3	2	1	2	1	..	1	8	6	6	...	..	...	..	...	...	4	1	...	11
	7	8	..	2	2	5	8	..	6	..	7	..	..	11	..	...	..	...	...	...	...	3	...
	8	..	3	5	1	..	..	..	..	..	..	..	..	...	..	...	..	...	...	...	...	...	...
	9	2	1	4	2	2	1	..	..	3	3	..	..	...	..	...	..	...	...	...	...	...	...
	10	..	..	..	..	..	..	..	..	..	..	..	..	...	..	...	..	...	...	...	...	...	...
	Total...	54	68	73	54	41	50	54	38	33	54	43	86	79	28	44	27	60	71	88	102	78	48

| | | |

action. Even after having been mineralized it may again be assimilated by fungi, algae, and bacteria.

However, more or less marked differences in the nitrogen availability become evident if the behavior of the various kinds of green manures is examined. Table 15 shows the percentage figures for the nitrogen returns in abridged form (without decimals). These figures exhibit clearly considerable differences among the green manures tested, and a summary of the initial and final effects (table 16) shows this even more definitely.

Pea and hairy vetch both gave a relatively large return in the first crop, but later the pea was outranked by the vetch. Horse bean acted differently according to its age: young material behaved much like pea and vetch, where-

TABLE 16

Percentage of nitrogen recovered in the first crop and in the following crops

POOR SOIL	PEA	HAIRY VETCH		HORSE BEAN		WHITE LUPIN			YELLOW LUPIN		
	Medium	Young	Medium	Young	Old	Young	Medium	Old	Young	Medium	Old
Crop 1	25	31	28	32	12	10	14	6	13	13	..
Crops 2–10	26	39	40	24	24	44	39	42	20	46	41
Total	51	70	68	56	36	54	53	48	33	59	41
	SWEET CLOVER			COWPEA					SOYBEAN		
	Young	Medium	Old	Young	Medium	Old	Ripe	Frozen	Young	Medium	Old
Crop 1	52	52	28	28	2	45	65	80	91	57	44
Crops 2–7	28	23	24	46	33	31	30	33	11	37	44
Total	80	75	52	74	35	76	95	113	102	94	88

as old material was similar to white and yellow lupines, which always acted little on the first, but more on the following crops. Sweet clover and soybeans, like horse beans, showed a slower and less complete effect with increased age, whereas mature cowpeas gave higher returns, especially after exposure to frost.

The physical and chemical characters of old horse beans, lupines, sweet clover, and soybeans explain their reduced efficiency. Their cellulose content increases considerably and the nitrogen content is reduced; accordingly, they stimulate microbial assimilation of nitrogenous compounds and reduce the mineralization of organic substances. Cowpeas, on the other hand, remain, even when their seeds are mature comparatively rich in nitrogen and low in cellulose content. Their high efficiency is, therefore, not very sur-

prising. Nevertheless, why freezing especially, has further stimulated this growing nitrogen efficiency can not be fully explained at present, whereas in the nitrification test the results have decreased with increasing age in this as in other cases. (Table 14.) The causes of the exceptionally low returns shown by young and medium aged cowpeas are entirely problematical. The regularity of their appearance precludes their being caused by an experimental error, and the similar inferiority exhibited by young yellow lupines when compared with older material indicates that they are caused by some unknown physiological peculiarities. Unsatisfactory results recorded occasionally with cowpeas and with yellow lupines in field tests may have been the outcome of similar conditions.

DISCUSSION

The experiments described on the preceding pages have confirmed earlier findings insofar as an average nitrogen return of approximately 50 to 60 per cent of the applied green manure could be recorded. However, only one-half to three-fourths of this amount was contained in the first crop; the residual effect was distributed in a regularly decreasing manner during the next two or three years. The separate findings showed wider fluctuations than have been recorded before. In the first crop they varied from negative results up to an apparent nitrogen availability of 149 per cent, whereas the total effects ranged from 33 to 208 per cent. It was shown that these high returns are due to a stimulation of the mineralization of humus nitrogen under the influence of relatively small applications of green manure, as are often used in the field. It is possible, therefore, that green manuring may sometime lead to a depletion in soil nitrogen, a fact which never before seems to have been noticed.

The biochemical transformations proceed more slowly in the field, than in pot experiments so that this undesirable influence upon the humus nitrogen is less likely to play an important rôle. But it is difficult, if not impossible, to decide in most cases, how far data obtained in field experiments actually represent the nitrogen availability of the green manures used. As a rule, the plants are plowed under where they have been grown. Thereby the specific effect exerted by the growing legumes upon soil productivity is more or less obliterated by the processes started in the soil by the incorporated green matter.

This specific effect, however, must be known in order to decide correctly whether the application of green manure is physiologically and economically sound. At first glance, it seems beyond doubt that the turning under of a green manure crop containing 100 pounds nitrogen per acre with an availability of about 50 per cent represents an efficient and economic means of improving the soil. Nevertheless a thorough knowledge of the action of the growing legumes as such, may change this view more or less.

Since under suitable conditions the nitrification of green manure nitrogen proceeds fairly rapidly during the first few weeks, it is easily understood why little, if any, noticeable effect can be expected if green manure is plowed under in summer or in fall while the soil is still warm and if the field is replanted several weeks or months later after heavy rains have washed out the nitrate which had been formed. Leguminous winter cover crops are generally much to be preferred, but in this case, too, it remains to be seen whether it is advisable to plow them under in spring or to use them in other ways.

SUMMARY

Field, greenhouse, and laboratory experiments upon the efficiency of green manures have furnished the following results:

(*a*) The nitrogen availability of green manures shows wide variations which are dependent on the quality and quantity of the green substances used and on the character of the manured soils. Small amounts of young materials, as a rule, give higher percentage returns than large quantities of old materials; but this rule is not without important exceptions. Cowpeas especially proved of greater value with increasing age, while young cowpeas, as well as young yellow lupins, showed an exceptionally low nitrogen availability. Frozen cowpeas displayed the highest efficiency.

(*b*) The average availability of green manure nitrogen was about 50 to 80 per cent if the green substances were added to the soil after they had been grown elsewhere and the tests were continued for several years. Similar results have been recorded repeatedly when green legumes were turned under where they had been grown. But it is an open question whether in such cases the effect is really the result of the green manuring, or whether it is more due to the influence of the growing legumes as such. The data recorded are in favor of the last named possibility.

(*c*) If green manures are incorporated into a soil not too poor in humus a general acceleration of the activities of the microörganisms living in the soil takes place with the result that the nitrification of the green manure nitrogen is accompanied by an intensified mineralization of the humus nitrogen. Accordingly, more nitrogen may be found in the first crop increases than has become available from the green manures. Occasionally on a rich soil more than 200 per cent of the nitrogen applied was returned within a few years. Nitrification tests made in the laboratory gave much lower figures than vegetation tests with this soil, whereas concordant results were obtained by both methods for a soil of low humus content.

REFERENCES

(1) Alway, F. J., and Bishop, E. S. 1911 Some notes on alfalfa and clover residues as sources of soil nitrogen. *Nebr. Agr. Exp. Sta. Ann. Rpt.* 25: 56–65.

(2) Arny, A. C., and Thatcher, R. W. 1915 The effect of different methods of inoculation on the yield and protein content of alfalfa and sweet clover. *Jour. Amer. Soc. Agron.* 7: 172–185.

(3) Bal, D. V. 1922 Studies on the decomposition of some common green-manuring plants at different stages of growth, etc. *Agr. Jour. India* 17: 133–151.

(4) Brigham, R. O. 1917 Assimilation of organic nitrogen by Zea Mays, etc. *Soil Sci.* 3: 155–195.

(5) Brown, P. E., and Stallings, J. H. 1921 Inoculated legumes as nitrogenous fertilizers. *Soil Sci.* 12: 365–407.

(6) CARR, R. H. 1916 Rate of humification of green manures. *Ind. Acad. Sci. Proc.* 1916: 398–402.
(7) COFFEY, G. N., AND TUTTLE, H. F. 1915 Pot tests with fertilizers compared with field trials. *Jour. Amer. Soc. Agron.* 7: 129–139.
(8) CRAIG, JOHN 1902 Orchard cover crops. *N. Y. (Cornell) Agr. Exp. Sta. Bul.* 198: 93–133.
(9) DUGGAR, J. F. 1898 Experiments with crimson clover and hairy vetch. *Ala. Agr. Exp. Sta. Bul.* 96: 181–208.
(10) DUGGAR, J. F. 1899 Winter pasturage, hay and fertility afforded by hairy vetch. *Ala. Agr. Exp. Sta. Bul.* 105: 127–160.
(11) DUGGAR, J. F. 1902 The cowpea and the velvet bean as fertilizers. *Ala. Agr. Exp. Sta. Bul.* 120: 121–179.
(12) FIPPIN, E. O. 1917 Livestock and the maintenance of organic matter in the soil. *Jour. Amer. Soc. Agron.* 9: 97–105.
(13) GRAUL, E. J., AND FRED, E. B. 1922 The value of lime and inoculation for alfalfa and clover on acid soils. *Wis. Agr. Exp. Sta. Res. Bul.* 54.
(14) GUILLIN, R. 1922 Dosage de l'azote assimilable dans les engrais. *Jour. Agr. Prat.* 86: 415–416.
(15) HART, E. B., AND BENTLEY, W. H. 1915 The character of the water-soluble nitrogen of some common feeding stuffs. *Jour. Biol. Chem.* 22: 477–483.
(16) HASELHOFF, E. 1924 Versuche über den Stickstoff haushalt im Ackerboden. *Landw. Vers. Stat.* 102: 73–89.
(17) HOFFMAN, M. 1911. Gründüngungswirtschaften. *Arb. Deut. Landw. Gesell.* 200: 64.
(18) HOPKINS, C. G. 1902 Alfalfa on Illinois soil. *Ill. Agr. Exp. Sta. Bul.* 76: 311–349.
(19) HUTCHINSON, C. M., AND MILLIGAN, S. 1914 Green manuring experiment, 1912–13. *Agr. Res. Inst., Pusa, Bul.* 40.
(20) JOSHI, N. V. 1919 Rate of nitrification of different green manures, etc. *Agr. Jour. India* 14: 395–409.
(21) JOSHI, N. V. 1922 Comparative manurial value of the whole plants and the different parts of green manures. *Agr. Res. Inst., Pusa,* Bul. 141.
(22) LANDER, P. E., WILSDON, B. H., AND MUKAND LAL, M. 1923 A study of the factors operative in the value of green manures. *Agr. Res. Inst., Pusa,* Bul. 149.
(23) LATHROP, E. C. 1916 Protein decomposition in soils. *Soil Sci.* 1: 509–532.
(24) LIPMAN, J. G., BLAIR, A. W., MCLEAN, H. C., AND WILKINS, L. K. 1914 Factors influencing the protein content of soybeans. N. J. Agr. Exp. Sta. Bul. 282.
(25) LÖHNIS, F. 1910 Handbuch der landwirtschaftlichen Bakteriologie, p. 578–582. Berlin.
(26) LUPTON, N. T. 1890 Pea vines as a fertilizer. Ala. Agr. Exp. Sta. Bul. 14.
(27) MARKLEY, K. S., AND HANN, R. M. 1925 A comparative study of the Gunning-Arnold and Winkler boric acid modifications of the Kjeldahl method for the determination of nitrogen. *Jour. Assoc. Off. Agr. Chem.* 8: 455–467.
(28) MARTIN, TH. L. 1921 Decomposition of green manures at different stages of growth. *N. Y. (Cornell) Agr. Exp. Sta. Bul.* 406: 139–169.
(29) MCBETH, I. G. 1917 Relation of the transformation and distribution of soil nitrogen. *Jour. Agr. Res.* 9: 183–252.
(30) MERTZ, W. M. 1918 Green manure crops in Southern California. *Calif. Agr. Exp. Sta. Bul.* 292: 1–31.
(31) MIELCK, OTFRIED. 1913 Die Wirkungen der Gründüngung. *Fühling's Landw. Ztg.* 62: 585–612.
(32) MITCHERLICH, E. A., KUNZE, R., CELICHOWSKI, K., AND MERRES, E. 1910 Ein Beitrag zur Düngemittel und Bodenanalyse. *Landw. Jahrb.* 39: 299–334.
(33) NOLTE, O. 1923 Gründüngung. *Flugschr. Deut. Landw. Gesell.* 23.

(34) PENNY, CH. L. 1903 Cover crops as green manures. *Del. Agr. Exp. Sta. Bul.* 60: 1–43.

(35) PENNY, CH. L. 1905 The growth of crimson clover. *Del. Agr. Exp. Sta. Bul.* 67: 1–53.

(36) POTTER, R. S., AND SNYDER, R. S. 1917 Decomposition of green and stable manure. *Jour. Agr. Res.* 11: 677–698.

(37) ROBERTS, I. P., AND CLINTON, L. A. 1897 Forage crops. *N. Y. (Cornell) Agr. Exp. Sta. Bul.* 135: 269–296.

(38) ROBINSON, C. S., AND WINTER, O. B. 1923 Some experiences with the alkaline permanganate method. *Jour. Off. Agr. Chem.* 5: 446–450.

(39) ROBINSON, T. R. 1908 The fertilizing value of hairy vetch for Connecticut tobacco fields. U. S. Dept. Agr. Bur. Plant Indus. Cir. 15.

(40) SCHNEIDEWIND, W. 1915 Die Ernährung der landwirtschartlichen Kulturpflanzen, p. 262. Berlin.

(41) SCHNEIDEWIND, W., AND MEYER, D. 1914 Über die Ergebnisse der in den Jahren 1911/13 . . . . ausgeführten Gründüngungsversuche. *Mitt. Deut. Landw. Gesell.* 29: 406–409.

(42) VORHEES, E. B. 1912 The use of green manures in soil improvement. *Cyclopedia of Amer. Agr.* ed. by L. H. Bailey 1: 503–509.

(43) WAGNER, P. 1915 Die Wirkung von Stallmist und Handelsdüngern. *Arb. Deut. Landw. Gesell.* 279.

(44) WHITE, J. W. 1918 Soil acidity as influenced by green manure. *Jour. Agr. Res.* 13: 171–197.

(45) WHITING, A. L. 1915 A biochemical study of nitrogen in certain legumes. Ill. Agr. Exp. Sta. Bul. 179.

(46) WHITING, A. L., AND SCHOONOVER, W. R. 1920 The comparative rate of decomposition of green and cured clover tops in soil. *Soil Sci.* 9: 137–149.

(47) WHITTLE, CH. A. 1920 Legume nitrogen. South. Fert. Assoc., Soil Impr. Com. Bul. 33.

(48) WIANCKO, A. T., FISHER, M. C., AND CROMER, C. O. 1914 Soybeans and cowpeas. Ind. (Purdue) Agr. Exp. Sta. Bul. 172.

(49) WRIGHT, R. C. 1915 The influence of certain organic materials upon the transformation of soil nitrogen. *Jour. Amer. Soc. Agron.* 7: 193–208.

WHY ARE SERPENTINE AND OTHER MAGNESIAN SOILS INFERTILE?

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General agreement exists among geologists and other students of soils respecting the marked inferiority as plant media, of soils derived from pure serpentine or from other highly magnesian rocks. No accord among scientists, has, however, been reached regarding the cause or causes of the infertility in question. The hypothesis which attempts to clarify the problem and to which many students of soils and plants have indicated their adherence, is briefly to the effect that the specific toxicity of magnesium accounts for the unsuitable nature of serpentine and similar soils as media for plant growth. Hilgard, among other investigators, accepted that hypothesis, particularly in view of Loew's well-known work on the rôle of calcium and magnesium in plant growth and on the lime-magnesia ratio. He added another view (2), however, to the effect that magnesian soils are usually deficient in "plant-foods" (4, 5).

It seemed to the authors that, inasmuch as the problem lends itself well to experimental test, it was high time to determine definitely the real answer to the question of the cause or causes of the infertility of serpentine soils. The following experiments, therefore, were planned and executed. Soils or freshly disintegrated serpentine rock was obtained from different locations in the hills of the Coast Range of the San Francisco Bay region, and submitted to the studies which are described below. The soils collected were as follows:

No. 1. From the west peak of Mount Tamalpais, Marin County. Disintegrating serpertine rock. No vegetation on it except an occasional tuft of moss on rock masses. Free from contamination with any other rock or soil.

No. 2. Vicinity of Mount Diablo, Contra Costa County. Taken from the top of a serpentine hill. No contaminating material. A very sparse grass vegetation existent.

No. 3. From same general location as No. 2, but at the foot of, and in a little vale between, two hills. Possibility of contaminating material. The soil was black, fine in texture, deep, and easily workable, thus distinctly different from the coarse material in No. 1 and No. 2. A fairly good grass vegetation existent, also scattered oaks and pine trees of fair vigor.

No. 4. From the side of a serpentine hill near Oakland, Alameda County. Disintegrated

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[1] Professor Bruce L. Clark, Associate Professor of Paleontology, University of California, was good enough to assist the authors in obtaining the samples of soil from the Mount Diablo region. For this assistance, the authors wish to express their sincere sense of obligation.

rock particles. No vegetation where sample was taken, but very sparse grass growth near place of sampling.

No. 5. Taken 3 feet from No. 4, among grass roots. The soil was only 6 inches deep.

No. 6. From a grassy knoll near No. 5. Only about one inch of soil and disintegrating rock. Grass very short and going to seed prematurely. Soil from among roots of grass.

No. 7. From among pine trees near No. 6. Soil about bare except for tree growth mentioned.

No. 8. From a location close to No. 4, 5, 6, and 7. Bottom of a knoll. Soil brown in color. A very poor growth of young eucalyptus trees.

No. 9. From a location near San Francisco. A black soil on disintegrated serpentine rock. Fairly good vegetation.

No. 10. From another spot in same location as No. 9. Disintegrated serpentine rock bearing a very poor vegetation.

COMPOSITION OF THE SOIL SOLUTION OF THE EXPERIMENTAL SOILS

Before proceeding to the determination of the cause of the well-known behavior of serpentine soils toward plants, it seemed highly desirable to learn something about the composition of their solution. To that end, a preliminary test was planned on a method to be employed for the purpose in subsequent work. The preliminary test consisted of obtaining the concentration

TABLE 1

Concentration of solution of Mount Tamalpais serpentine soil

	C_2H_5OH DISPLACEMENT	ACETONE DISPLACEMENT	WATER EXTRACT 1:1
Total solids, *p.p.m.*	144	150	166
Freezing point depression, °*C.*	0.021	0.021	0.026

of the solution of three samples of the Mount Tamalpais rock powder by the following methods:

(*a*) Parker's displacement method with acetone, (*b*) Parker's displacement method with alcohol, (*c*) water extract, using 1 part soil to 1 part water, and (*d*) freezing point depression of solutions obtained by the three foregoing methods, in addition to the gravimetric determinations upon them.

Table 1 gives the results obtained on the three soil samples in question. Only an average of the values obtained is given, since agreement between the samples was very good.

Even the foregoing simple preliminary tests gave us a significant hint of the answer to our problem in the remarkably low concentration of the soil solution, but it seemed at once desirable to have more detailed data on the composition of these soil solutions. Inasmuch as there were no significant differences between the values given in table 1 as representing different methods, the water extract method was used in the subsequent determinations. In addition to analyzing the soil extracts thus obtained, however,

conductivity determinations were made on the same solutions, and the pH was determined throughout. Briefly, the following procedure was employed in the analyses of the ten samples of soil described above. The soil was passed through a 1-mm. sieve. It was then shaken with an equal weight of distilled water for one hour and a half in a mechanical shaker. The extract was filtered through a Pasteur-Chamberland filter and analyzed by the standard methods in vogue in modern laboratories. The nitrate content of the extract was determined on two soils by two methods so as to check on the reliability of the data. The methods used were the phenol-disulfonic acid method and the Devarda method. The two methods checked to within one part per million. The conductivity of the soil extracts was determined by the standard methods. The pH values were determined electrometrically,

TABLE 2

Analysis of serpentine soils

(In parts per million)

SOIL NUMBER	pH VALUE	TOTAL NITROGEN	NO_3	CONDUCTIVITY SPECIFIC RESISTANCE	TOTAL SOLIDS	NON-VOLATILE SOLIDS	Ca	Mg	HCO_3	PO_4
				ohms						
1	8.20	?	Trace	..	166	92	20	21	150	Trace
2	8.12	140	13	3,336	324	157	20	22	20	4
3	8.12	115	8	4,200	247	145	14	31	18	..
4	8.18	78	2	2,291	..	..	16	20	21	..
5	8.01	112	2	2,957	..	..	..	..	23	..
6	5.98	286	6	1,888	..	..	..	..	7	..
7	7.90	50	2	4,308	250	117	..	..	16	..
8	8.10	133	3	3,052	358	208	11	22	19	..
9	7.65	136	2	2,176	450	210	19	45	18	..
10	8.11	96	2	2,232	..	..	..	..	12	..

using 10 gm. of soil and 25 cc. of CO_2-free water for every soil. The results of all these analytical studies are set forth in table 2.

The results of the analyses as submitted in table 2 are interesting even at a casual glance. In the first place, they check the freezing point determinations in testifying to the strikingly low concentration of solutes in the material in question. In the second place, the data for pH determinations show with only one exception that the soils or rock powders are decidedly alkaline. In the third place, the conductivity measurements stated in ohms as specific resistance check the freezing point determinations as well as the analyses. Finally, the lack of PO_4 ion in all solutions but one testifies to the effectiveness of the alkalinity in removing phosphorus from the soil solution. The values for Ca and Mg are not striking except to indicate that the magnesium content cannot well be the dominant factor in determining the sterility of

the soil in question. It is, perhaps, worthy of special comment that all the soil and rock powders studied are shown in table 2 to possess very small quantities of nitrate. Further comment with respect to this aspect of our problem will be made below.

Although the quantity of volatile matter in the soils is not high, it should be remarked that when the serpentine materials were dried at 100°C., they suffered losses of 30 to 40 per cent in weight and were difficult to bring to constant weight. This indicates a probable very high content of combined water in the molecules making up the rock material.

PHYSIOLOGICAL EXPERIMENTS

In view of the interesting analytical data briefly discussed above, the authors decided upon a program of experimentation in cultures which might be calculated to determine the validity or invalidity of the conclusions suggesting themselves from table 2, which are as follows:

1. Magnesium is probably not responsible for the sterility of serpentine soils.
2. The high pH of the soil solutions in question is a suspicious factor in the problem.
3. The very low content of important ions in the solutions, and particularly the NO_3 and PO_4 ions, constitute another suspicious factor in the problem.

In the culture experiments, barley was grown in extracts of some of the soils in question, and in artificially compounded solutions of similar composition. The serpentine soils at first chosen for making extracts were the Mount Tamaplais and the Mount Diablo soils since both gave chemical characteristics strikingly similar, as evidenced in table 2, and yet were strikingly dissimilar in appearance, as the descriptions given above will readily make clear.

The barley plants were grown, therefore, in culture solutions placed in fruit jars of approximately one liter capacity. For each kind of solution, jars were used in quadruplicate, and 5 plants were grown in every jar, thus 20 plants for each type of medium allowed some control of individual variation among the plants. To serve as a control culture solution for comparison with the soil extracts and with other similar solutions, a medium composed of 0.004 *M* $MgSO_4$, 0.004 *M* $Ca(NO_3)_2$, and 0.004 *M* KH_2PO_4 and with about 0.5 atmosphere osmotic pressure, was employed. In cases in which two salt constituents were added in the test solutions, the total quantity used was such as to be equal to that of the foregoing control solution, namely, 0.012 mol of salts per liter. In other cases in which but one salt was added, a quantity of the salt equal to 0.005 mol per liter was employed in order to keep the individual ionic concentrations approximately comparable with those of the control solution. In the natural soil extracts, of course, the concentrations of solutes were lower than in the artificially prepared or modified solutions, as exemplified in the Mount Tamalpais soil, which has a concentration equivalent to less than 0.005 mol per liter. The solutions were not changed during

the culture period, but distilled water was added from time to time to make up for losses by transpiration, and $FeSO_4$ was added as needed.

For a clearer presentation of the data on the culture experiments, the soils tested will be considered separately.

The Mount Tamalpais soil

Since it was difficult to obtain large quantities of this soil for preparation of soil extracts, only three series of culture solutions were prepared therewith. The other eight series were made up artificially to be identical with the first three. The kinds and amounts of salts necessary for the artificially prepared solutions were determined by calculating the reaction values of the salts of the original solution from which the molal concentration may be calculated.

TABLE 3

Analysis of the Mount Tamalpais soil

IONS	QUANTITY FOUND	EQUIVALENT WEIGHT	REACTION VALUE	CONCENTRATION
	p.p.m.			*mols.*
Na	12.0	23.0	0.52	0.00052
K	6.4	39.0	0.17	0.00017
Mg	21.0	24.3	1.72	0.00172
Ca	20.8	40.7	1.02	0.00102
			3.43	0.00343
CO_3	13.0	76.3	0.34	0.00034
SO_4	5.4	96.0	0.11	0.00011
Cl	15.0	35.5	42.0	0.00042
HCO_3	153.0	61.0	2.52	0.00252
			3.39	0.00339

The reaction value in each case is found by dividing the quantity in parts per million found by the equivalent weight. The method is made clear in table 3.

The plants in the Mount Tamalpais series were grown for a little over five weeks in the fall of the year. The results obtained are recorded in table 4.

Because of an accident, the plants in the Mount Tamalpais series were lost before their dry weights could be determined. Table 4 shows, however, the lengths of tops and roots obtained in the several cultures. It is clear from the data in table 4 that two factors limit the growth of plants in extracts from the Mount Tamalpais soil. One is the high pH and the other is the lack of NO_3 and PO_4 ions. In this case, there is a smaller effect resulting from the correction of the pH than from the addition of necessary ions. Nevertheless the correction of the pH is clearly effective when the NO_3 and PO_4 ions are not limiting factors, as a comparison of series IV with series II indicates. These data would seem to give tangible support for the conclusions drawn from the analytical data submitted in table 3.

TABLE 4

Data on growing barley in the aqueous extract of the Mount Tamalpais soil (soil No. 1) and in similar solutions

JAR NUMBER		COMPOSITION OF SERIES											
		No. I	No. II	No. III	No. IV	No. V	No. VI	No. VII	No. VIII	No. IX	No. X	No. XI	No. XII
		Control 0.5 atm.	Soil extract Na_2HPO_4, NaH_2PO_4, KNO_3 +++	Soil extract + HCl	Soil extract NaH_2PO_4 KNO_3 HCl +++	Soil solution	Soil solution + Na_2HPO_4	Soil solution + KNO_3	Soil solution KNO_3 $NaHPO_4$ ++	Soil solution + KCl	Soil solution $MgHPO_4$ HCl ++	Soil solution $MgHPO_4$ KNO_3 HCl +++	Soil solution $Ca(NO_3)_2$ $Mg(NO_3)_2$ HCl +++
1	Tops, *cm*.	55	45	17	55	20	0.15	45	48	17	18	51	43.18
	Roots, *cm*.	25	32	20	27	25	53	20	33	30	52	25.4	24.4
2	Tops, *cm*.	53	42	20	55	17	15	47	44	18	17	51	41
	Roots, *cm*.	27	30	30	27	15	50	20	30	33	54	25.5	25.9
3	Tops, *cm*.	53	43	17.5	62	20	13	46	40	18	19	52.1	42
	Roots, *cm*.	25	27	30	23	20	58	20	35	33	54	26	25.8
4	Tops, *cm*.	48	43	15	47	18	13	47	37	15	17	49.7	39.8
	Roots, *cm*.	23	30	22.5	26	20	60	20	30	30	56	25.8	25.2
pH value at beginning of experiment		..	8.1	6.0	6.0	8.2	8.1	8.1	8.1	8.0	5.2	5.2	6.0
Average length of tops, *cm*.		52	43	17	55	19	14	47	42	17	18	51	41
Average length of roots, *cm*.		25	31	25	26	20	55	20	32	32	54	27	25

TABLE 5

Data on growing barley in the aqueous extract of the Mount Diablo (soil No. 2) and in similar solutions

DESCRIPTION	COMPOSITION OF SERIES									
	No. I	No. IA	No. II	No. III	No. IV	No. V	No. VI	No. VII	No. VIII	No. IX
	Control 0.5 atm.	Control 0.5 atm.	Soil extract	Soil extract + KNO_3	Soil extract + $NaNO_3$	Soil extract + HCl	Soil extract + KNO_3 + + HCl	Soil extract + $NaNO_3$ + + HCl	Soil extract + KNO_3 + + NaH_2PO_4	Soil extract + KNO_3 + + Na_2HPO_4
pH value before growing	7.2	5.5	8.1	8.1	8.1	5.5	5.5	5.5	5.5	8.1
Length of tops, *cm*	65	64	25	42	40	24	48	38	78	45
Length of roots, *cm*	25	25	75	45	28	40	35	55	30	28
Average weight of tops, *gm*	4.86	3.72	0.53	1.67	1.44	0.61	2.98	1.95	6.39	2.25
Average weight of roots, *gm*	1.14	1.29	0.24	0.81	0.40	0.30	1.10	0.50	1.52	0.58
Inflorescences	30	30	1–3	7	10–20	1–3	18	12	30	18
Average of total weight, *gm*	4.5	5.06	0.79	2.6	1.88	0.91	3.83	2.33	7.77	2.50

The Mount Diablo soil

At this juncture, it seemed essential, however, to obtain more extensive and complete data with other serpentine soils. An experiment similar to the one just described, therefore, was started with extracts of one of the Mount Diablo soils (No. 2) instead of the Mount Tamalpais soil. The results are given in tables 5 and 6.

The data in tables 5 and 6 not only confirm, but considerably strengthen those of table 4. The additional evidence furnished in the values for the dry weight of the plants and in the number of heads produced gives conclusive proof that the magnesium content of the serpentine soil extract is not the cause of the depression of plant growth therein. On the other hand, they confirm

TABLE 6

Weight of tops and roots of barley plants grown in the solutions given in table 5

JAR NUMBER	DESCRIPTION	SERIES									
		No. I	No. IA	No. II	No. III	No. IV	No. V	No. VI	No. VII	No. VIII	No. IX
		gm.	*gm.*	*gm.*	*gm.*	*gm.*	*gm.*	*gm.*	*gm.*	*gm.*	*gm.*
1	Tops	4.93	3.80	0.54	1.67	1.47	0.64	2.98	1.82	6.33	2.08
	Roots	1.07	1.43	0.23	0.80	0.41	0.30	1.00	0.47	1.33	0.50
2	Tops	4.79	3.60	0.49	1.72	1.48	0.57	2.90	1.94	5.87	2.80
	Roots	1.00	1.21	0.25	0.79	0.40	0.26	1.03	0.51	1.46	0.68
3	Tops	4.80	3.72	0.58	1.62	1.36	0.64	3.03	2.13	6.63	2.03
	Roots	1.20	1.18	0.26	0.78	0.42	0.30	1.24	0.52	1.65	0.60
4	Tops	4 91	3.80	0.52	1.67	1.52	0.58	3.02	1.92	6.77	2.10
	Roots	1.30	1.35	0.23	0.85	0.38	0.35	1.13	0.50	1.65	0.53
Average of total weight....		4.50	5.06	0.79	2.60	1.88	0.91	3.83	2.33	7.77	2.50

the conclusion, which may be drawn from the length of tops and roots yielded in the cultures, that the high pH of the solutions and the deficiency first in NO_3 ion and secondly in PO_4 ion, limit the growth of plants in those solutions. This evidence is the more striking since the Mount Diablo soil is totally different in appearance and age from the Mount Tamalpais soil which it resembles so strongly in its physiological effects. The more subtle factors common to serpentine soils and not their outward appearances are, therefore, their dominant features from the physiological standpoint.

In order to strengthen the evidence, the results of an experiment with still another Mount Diablo soil (No. 3) conducted as a parallel with soil No. 2 are given in tables 7 and 8.

Again, there is evidence in a soil of very different appearance from either of the foregoing, of the existence of a common and dominant set of factors

determining their physiological value. (Tables 7 and 8.) Again, we have striking evidence of the effectiveness of the factors common to serpentine

TABLE 7

Data on growing barley in the aqueous extract of the Mount Diablo soil (soil No. 3) and in similar solutions

DESCRIPTION	COMPOSITION OF SERIES								
	No. XI	No. XII	No. XIII	No. XIV	No. XV	No. XVI	No. XVII	No. XVIII	No. XIX
	Soil extract	Soil extract + KNO_3	Soil extract + $NaNO_3$	Soil extract + HCl	Soil extract + KNO_3	Soil extract + $NaNO_3$	Soil extract + NaH_2PO_4 + KNO_3	Prepared solution 0.003 mol. (page 298)	Soil extract + KNO_3 + Na_2HPO_4
pH value before growing	8.2	8.2	8.2	5.5	5.5	5.5	5.5	7.3	8.2
Length of tops, *cm*.	30	38	34	28	42	40	82	29	70
Length of roots, *cm*.	90	60	62	75	60	56	27	34	27
Average weight of tops, *gm*.	0.79	1.50	1.28	0.72	2.15	1.83	7.37	0.83	4.28
Average weight of roots, *gm*.	0.32	0.68	0.35	0.30	0.85	0.42	1.64	0.27	1.22
Average of total weight, *gm*.	1.10	2.15	1.62	1.04	2.95	2.34	9.01	1.08	5.50
Inflorescense	1–3	6	5	3	8–10	6	30	3–5	30

TABLE 8

Weight of tops and roots of barley plants grown in the solution given in table 5

JAR NUMBER	DESCRIPTION	SERIES								
		No. XI	No. XII	No. XIII	No. XIV	No. XV	No. XVI	No. XVII	No. XVIII	No. XIX
		gm.	*gm.*	*gm.*	*gm.*	*gm.*	*gm.*	*gm.*	*gm.*	*gm.*
1	Tops	0.73	1.67	1.44	0.80	2.10	1.82	7.29	0.84	4.41
	Roots	0.29	0.68	0.36	0.30	7.50	0.40	1.70	0.20	1.34
2	Tops	0.78	1.38	1.17	0.77	2.23	1.84	7.43	0.82	4.32
	Roots	0.32	0.57	0.30	0.27	0.78	0.42	1.80	0.28	1.29
3	Tops	0.74	1.60	1.37	0.62	2.26	1.77	7.21	0.86	4.27
	Roots	0.26	0.72	0.42	0.30	0.98	0.40	1.50	0.23	1.13
4	Tops	0.92	1.33	1.13	0.75	2.00	1.87	7.48	0.82	4.12
	Roots	0.42	0.64	0.31	0.35	0.89	0.45	1.56	0.28	1.10
Average of total weight		1.10	2.15	1.63	1.04	2.95	2.35	9.00	1.08	5.50

soils and again, there appears to be no evidence that the magnesium content of the soil extracts is the toxic, inhibiting, or dominant factor.

In view of the decisive nature of the foregoing results, it seemed unnecessary to test the solutions of the other serpentine soils which had been collected as above described.

GENERAL DISCUSSION

There is a striking agreement between the general effects of the sepentine soil solutions, regardless of the apparent differences between the soils as regards their effects on the growth of barley. The minor differences in their effects as noted in the tables are easily explained on the basis of the different periods of the year in which the several experiments were conducted and on the different periods for which the cultures were grown. This would seem to indicate that the effects of the solutions in question on barley plants are not explicable on any irregular or accidental constituents of serpentine soils, but on constant and dominant characters.

Our data show, further, that there is confirmation in the conditions found (*a*) in the native vegetation on serpentine soils, (*b*) in the kind of plants grown on soil extracts from them and on artificial solutions made to simulate them in composition, and (*c*) in the analytical data showing the outstanding characters of the serpentine soils.

The small amount of magnesium found in all the serpentine soil extracts when viewed in the light of the results obtained by Gericke who grew plants successfully in solutions whose salt constituents were seven-eighths magnesium salts, would, in itself, give no ground for attributing the partial or complete sterility of serpentine soils to that factor. But when it is realized, as the foregoing experiments compel one to do, that the addition of a lacking ion or a change in the pH, or both, in a serpentine soil extract with the same magnesium content, changes it from a poor to a good medium for a barley plant, a full answer is given to the question which forms the subject of this paper. In fact the authors have even added magnesium to some of their culture solutions in the foregoing experiments without rendering them toxic or more toxic thereby.

It is not the magnesium content but the high pH value which is the dominant and consistent characteristic of serpentine soil extracts. It is known, of course, that the weathering of serpentine rock results in a mixture of carbonates and silicates. These must give rise to the HCO_3 and SiO_3 ions found on analysis in the serpentine soil extracts which, in turn, on hydrolysis give the high pH values above shown. At equilibrium between the serpentine soil and its solution, the pH value is about 8.1. In growing on such a soil, a plant would tend to lower the pH, but this would result in further hydrolysis of the compounds in question with resultant maintenance of a high pH. It is only in this very indirect sense that Mg salts in serpentine soils can be said to be responsible for their infertility.

The very dilute solution which characterizes serpentine soils is worthy of special comment. The authors have found a very small colloid content in

all the serpentine soils. This may, perhaps, account in some measure for the lack, of retentiveness of important ions for plants by the soils in question, but it is more likely that the unique chemical character of the rocks contributing to such soils is a more important factor in accounting therefor. These considerations would seem to offer the key to the solution of the problem of rendering serpentine soils more fertile.

SUMMARY AND CONCLUSIONS

1. A study was made of 10 widely different serpentine soils from different localities.

2. All of the soils were analyzed and the conductivity of their solutions was determined. It was found that:

All soils were poor in available important ions.
The nitrate content was low even where the total nitrogen was considerable.
Most of the soils had a pH value generally of about 8.1.
Although serpentine soil is derived from a magnesian rock, the ratio of magnesium to the total concentration of the soil extract was not necessarily high.
A few of the soil extracts were analyzed for phosphate and potassium, and were found to be deficient in these constituents.

3. Three of the soils were studied by growing barley in culture solutions (prepared either by adding certain constitutents to their soil extracts, or to solutions like their soil extracts, but artificially prepared.) It was found that:

The addition of nitrate improved the growth of the plants.
The lowering of the pH value improved the plant growth where necessary ions had been added.
The addition of potassium improved the root development of the plants.
The addition of phosphate increased the growth of the plants.
The addition of magnesia to soil solutions from one soil had no effect on the growth of the barley.
The growth of the plants was at its maximum when several essential ions were added and the pH value was lowered.

4. The experiments with barley also showed, like the analytical data, that the soil extracts were poor in nitrate, phosphate, and potassium, chiefly the first two.

5. There is a strong parallelism between the composition of serpentine soils and the vegetation found on them as well as the behavior of barley grown in extracts from them.

6. The infertility of serpentine soils is not caused by a too high content of soluble magnesium, but by a high pH and a deficiency in certain ions, chiefly nitrate and phosphate. This conclusion reverses all the teaching on this subject heretofore given, especially that as regards the toxicity of magnesium in serpentine soils.

REFERENCES

(1) BLACKSHAW, G. N. 1921 Magnesia impregnated soils. *So. African Jour. Sci.* 17: 171–78.

(2) HILGARD, E. W. 1906 Soils, p. 36. New York.

(3) LIPMAN, C. B. 1916 A critique of the hypothesis of the lime-magnesia ratio. *Plant World*, 19: 88–105.

(4) LOEW, O. 1901 The relation of lime and magnesia to plant growth. I. Liming of soils from a physiological standpoint. *U. S. Dept. Agr. Bur. Plant Indus. Bul.* 1: 9-35.

(5) MAY, W. D. 1901 The relation of lime and magnesia to plant growth. II. Experimental study of the relation of lime and magnesia to plant growth. *U. S. Dept. Agr. Bur. Plant Indus. Bul.* 1: 37–53.

(6) PARKER, F. W. 1921 Methods of studying the concentration and composition of the soil solution. *Soil Sci.* 12: 209.

THE TOLERANCE OF PLANTS FOR NaCl

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A number of circumstances, economic and scientific, have conspired to invest the subject of salt effects in plant growth with outstanding importance in the realm of plant physiology. When we remember that the root medium in nature is a weak salt solution whose changing composition within a season is an important determinant of plant growth, that the introduction of salts into that medium may exert marked influences on the absorptive capacities of the plant for necessary and unnecessary ions, that large concentrations of sodium salts and others may depress and even exterminate plant growth, that the resistance of plants to such baneful effects of excessive quantities of salts may lead to important physiological and genetic discoveries—we have before us a few of the many cogent reasons for studying systematically and intensively the relations of salts to plant growth.

Such were the considerations which induced the authors to undertake to contribute another chapter, in addition to those already contributed by one of the authors, to the story of salt relations of plants. The aim in these experiments was to determine the tolerance of plants for NaCl under extreme conditions, to note if any stimulating effects ever enter, and to discover if different kinds of plants react differently to the same concentration of the salt.

PLAN OF EXPERIMENT

In formulating the procedure to be adopted in their experiments, the authors decided that all soil culture experiments on the subject of the toxicity of NaCl or of the tolerance of plants for NaCl are seriously at fault, in that uncontrollable factors or at least unknown factors are introduced into the experiment by the use of soils as media. Both culture solution experiments and sand culture experiments are on record in sufficient number to show that soil and possibly other solid media do not lend themselves well to the prosecution of research on the tolerance of plants for salts. In the case of soils particularly, the well-known phenomenon of the replacement of bases which follows upon the heels of the introduction of any salt like sodium chloride into the soil, is likely to bring about a condition which masks the real nature of the effects of a salt upon a plant. At this juncture in the development of the science of plant physiology and its associated sciences, the phenomena just

referred to, including the replacement of bases and the antagonism between ions, are so thoroughly appreciated by plant physiologists as to require no reference to the long list of articles which testify to the interest they have stimulated.

In view of what has been said, we determined to employ the culture solution method in the present studies. Although, to be sure, the constituent salts used in the culture solution do exert some effect upon the salt whose specific relations with the plant are under investigation, such effects cannot well be considered as being of great magnitude inasmuch as the sodium chloride under study in our work is used almost throughout in such large concentrations as to overshadow in its effects any neutralizing reactions which other salts present in the medium with it may exert thereon. The basic culture solution which was used in these experiments had the following composition in grams per liter, the water of crystallization being included:

$Ca(NO_3)_2$	1.721
KNO_3	0.321
KH_2PO_4	0.249
$MgSO_4$	0.993

The ionic composition of this solution in parts per million was as follows:

Ca	292
NO_3	1100
Mg	98
PO_4	174
SO_4	372
K	194

To this solution there was added throughout, four drops of a 5 per cent solution of ferrous sulfate to each 2-liter jar. Similar amounts of iron were added whenever the condition of the plants indicated a need. In each 2-quart Mason jar, 1700 cc. of the nutrient solution just described was placed. After having introduced into the jar a sufficient amount of concentrated NaCl solution to make up the desired concentration of NaCl, the total amount of the solution was brought up to 1900 cc. in every jar. The plants were set into the paraffine cork stoppers after the usual fashion employed in such experiments. Every concentration of salt was tested in quadruplicate. In the case of wheat and barley, five plants were set in each jar; in the case of peas, only three plants. All the plants were grown in a greenhouse where the temperature was not controlled but varied within a considerable range.

RESULTS OF THE WHEAT SERIES

The concentrations of NaCl employed in the wheat series varied from 500 p.p.m. to 15,000 p.p.m. with intervals of 500 p.p.m. between each of two successive sets of jars. The wheat seedlings were set out in December, 1920. In the second week of their growth in the jars, it was possible to detect dif-

ferences in the heights of the tops of the plants growing in concentrations of NaCl above 8500 p.p.m. In the concentrations below that, the tops seemed to be of uniform height throughout. No growth, as judged by increase in length, was detected in concentrations of NaCl above 14,000 p.p.m., but at this time all the plants were still alive. As regards the root development during the second week, it was possible to notice two maxima in root length. The roots in the control set of jars which received no sodium chloride were normal, those growing in 500 p.p.m. were only about three-fourths the length of the former; and those at higher concentrations of NaCl were showing a gradual lengthening over those in the 500 p.p.m. concentration until a concentration of 2500 p.p.m. was reached, where the roots appeared to be as long as they were in the control jars. Beyond 4000 p.p.m. there was a gradual decrease in the length of the roots until a concentration of 14,000 p.p.m. was reached, beyond which there was no growth evident. The roots even at this early stage were beginning to disintegrate, as shown by a slimy coating on their surfaces.

Four weeks after planting, the tops of the plants at concentrations of 6500 p.p.m. and above were noticeably thinner and a little shorter than at any concentration below. Even at this date there was little difference in height among the plants in all the sets up to 6500 parts NaCl per million. The tops of the plants all appeared to be healthy even in the highest concentration of 15,000 p.p.m. where no growth was taking place. Nevertheless, the gradual disintegration of the plants in the high concentrations of sodium chloride was visible at this point. In the root systems at the same date, the two maxima for root length and development which were noted above had become accentuated so that the roots in a concentration of 500 parts NaCl per million were no more than one-half as long as those in the control. Those in 1000 p.p.m. were about the same length as those in 500 p.p.m. Then, as the concentrations of NaCl increased, the length of the roots increased gradually up to 3000 parts NaCl per million, where the roots were slightly longer than those of the controls. From 4000 p.p.m. NaCl to higher concentrations there was a gradual decrease in the root length, the roots becoming more and more stubby, and slightly brown and discolored above 8000 p.p.m. NaCl.

At the third observation, about two weeks subsequent to the one just described, the color of the plants was good in all the sets except those of 15,000 p.p.m. NaCl. A few jars in which higher concentrations of NaCl than 15,000 p.p.m. were employed contained no living plants, and only a few plants were alive in concentrations above 14,000. In the lower concentrations, the vigor of the plants was clearly manifest. The plants tillered well and there seemed to be little difference between the control plants and those growing in concentrations of NaCl up to 4,000 p.p.m. Beyond the latter concentration the plants gradually decreased in thickness of stalk and width of leaf. Nevertheless, the plants growing in concentrations of 10,000 p.p.m. NaCl appeared normal when viewed alone. The roots appeared to be normal, and only a comparison of these plants with the control showed the latter to be superior.

The roots at this observation were long, white, and branched in the control cultures. The roots in 500 p.p.m. NaCl were markedly dwarfed and showed a tendency to send out long, thick, unbranched roots from the crown. This was true in all the four jars of this same concentration. Again it was noticed, as on the previous occasion, that the root system gradually increased in length with the increase of concentration of NaCl up to 4,000 p.p.m., where they were equal in length to those of the control. There was a slight difference between these and the control roots in other respects, however, in that they

TABLE 1

Tolerance of wheat plants for NaCl

Dry weight per culture (5 plants)

CONCENTRATION NaCl	TOPS						ROOTS	
	1	2	3	4	Total	Average	Total	Average
p.p.m.	*gm.*	*gm.*	*gm.*	*gm.*	*gm.*	*gm.*	*gm.*	*gm.*
000	11.8	11.8	11.6	9.3	44.5	11.1	6.9	1.70
500	13.8	14.8	13.8	16.6	59.0	14.7	9.8	2.45
1,000	14.1	12.3	14.9	14.9	56.2	14.1	8.6	2.15
1,500	18.8	15.4	15.5	16.1	65.8	16.4	10.1	2.50
2,000	16.1	15.7	16.4	15.9	64.1	16.0	10.3	2.56
2,500	18.0	13.0	17.5	14.9	63.4	15.8	9.6	2.40
3,000	15.7	18.7	21.6	17.3	73.3	18.4	11.2	2.80
3,500	16.0	17.5	16.5	17.9	67.9	17.0	9.6	2.40
4,000	12.4	17.4	21.0	17.7	68.5	17.1	10.5	2.60
4,500	16.3	17.9	17.0	18.1	69.3	17.3	10.6	2.65
5,000	20.6	21.0	18.4	19.6	79.6	19.9	12.2	3.00
5,500	14.7	11.0	15.8	14.3	55.8	14.0	9.1	2.30
6,000	19.9	18.3	21.6	9.7	59.5	14.9	9.0	2.30
6,500	17.5	12.1	11.1	18.2	58.9	14.7	9.8	2.50
7,000	7.2	14.8	15.3	18.9	56.2	14.1	8.5	2.10
7,500	17.0	16.0	11.5	13.3	57.8	14.4	9.5	2.40
8,000	5.1	10.2	11.8	13.3	40.4	10.1	6.1	1.50
8,500	7.0	6.7	8.2	10.2	32.1	8.0	5.1	1.30
9,000	6.3	6.0	6.2	6.9	25.4	6.3	4.6	1.20
9,500	8.4	4.5	4.0	1.9	18.8	4.7	5.4	1.40
10,000	5.9	4.3	0.6	1.9	12.8	2.5	2.2	0.55

were slightly thicker, the branches were shorter, and the color slightly browner. Beyond 4000 p.p.m. concentration up to 14,000 p.p.m., there was a steady falling off in length in the root systems. In concentrations between 7000 and 12,000 p.p.m. NaCl the root branches were short, allowing the roots to be readily separated from one another.

Two weeks following the third series of observations (8 weeks after planting), the tops and the roots of the plants both showed differences in length over those noted earlier. There seemed to be at this point a gradual increase in height of the tops as one examined the concentrations from 500 to 4500 p.p.m.

NaCl. The control plants were still better than those in 500 p.p.m. NaCl. The maximum height of the tops was at a concentration of 4500 p.p.m. at this point in the growth of the plants. Beyond 13,000 p.p.m., all plants were dead. In the case of the roots, the differences in length which were so marked two weeks previously were at this date not so great, the tendency being seemingly in the direction of an equalization in the length of the roots in all the series up to 4500 p.p.m.

The last observations were made when the plants were about nine weeks old and only a few days before harvesting. The differences in length of root which are described above had almost entirely disappeared. Nevertheless, the roots of the plants in concentrations of 500 p.p.m. NaCl were still the shortest of the whole series except those in which distinct injury had occurred, namely in concentrations of 7500 p.p.m. or above.

The experiment was terminated about March 10, 1921. The plants were all dried and weighed. The roots were separated from the tops and, in order that the data might be subjected to statistical study, the top of each plant was weighed separately. Tables 1 and 2 give the data obtained on the dry weights of the plants which were harvested as follows: table 1 gives the results of the total weights of tops and roots from each jar, and table 2 gives the weight of each plant top separately. It is perfectly clear from these data that when tested by the solution culture method, NaCl does not become toxic to the wheat plant when grown under the conditions described until a concentration of approximately 8000 p.p.m. is reached, and at all concentrations of NaCl below 8000 p.p.m., the growth of the tops and roots, and particularly the former, is markedly stimulated by NaCl. When the yields of dry matter of individual plants or of groups of five plants together in every jar are compared with those of the control cultures in which no NaCl was included, but which were otherwise entirely suitable for the growth of the wheat, as the yields in the control cultures indicate, it is apparent that these are not accidental occurrences. It is also apparent that at 8500 p.p.m. and above, NaCl becomes very toxic and depresses the growth of the wheat plant until at 10,000 p.p.m., only about one-fifth of the dry weight of the control cultures is obtained. The break in the curve on dry weight yields is a fairly sharp one beyond the concentrations of 7500 p.p.m. NaCl. The dry weights of the roots parallel fairly closely those of the tops, so far as the important breaking point in the curve is concerned, at any rate. It is evident from these results that the lethal concentration of NaCl for the wheat plant under the conditions in question is somewhere in the vicinity of 10,500 p.p.m. when plant growth for the whole normal life period is considered. It is probably much higher than that for a very short period of say from two to four weeks of the life of a plant, as the observations above indicate.

All this is very different from the usual conception of the relation of common salt to the growth of the wheat plant. The fact that NaCl, under certain conditions, will markedly stimulate the growth of the wheat plant at all

TABLE 2

Tolerance of wheat plants for NaCl

Dry weight of individual tops

CONCENTRATION NaCl	JAR 1					JAR 2					JAR 3					JAR 4					MEAN DRY WEIGHT TOPS	DRY WEIGHT OF ROOTS				AVERAGE DRY WEIGHT OF ROOTS
																						Jar 1	Jar 2	Jar 3	Jar 4	
p.p.m.	*gm.*	*gm.*	*gm.*	*gm.*	*gm.*	*gm.*	*gm.*	*gm.*	*gm.*	*gm.*	*gm.*	*gm.*	*gm.*	*gm.*	*gm.*	*gm.*	*gm.*	*gm.*	*gm.*	*gm.*	*gm.*	*gm.*	*gm.*	*gm.*	*gm.*	*gm.*
000	2.6	2.0	1.9	1.7	3.6	2.9	1.7	2.4	2.5	2.3	3.0	2.2	2.4	1.7	2.3	2.0	2.7	1.3	1.6	1.7	2.23 ± 0.08	2.0	1.6	1.8	1.5	1.7
500	2.2	3.6	3.2	2.3	2.5	2.2	3.4	3.1	3.4	2.7	2.6	2.9	2.5	2.9	2.9	5.3	3.4	2.1	3.3	2.5	2.96 ± 0.10	2.6	2.4	2.0	2.8	2.5
1,000	2.4	3.2	3.1	2.7	2.7	2.5	1.4	3.8	1.5	3.1	2.5	2.2	2.9	2.9	4.4	3.7	2.7	2.9	2.6	3.0	2.81 ± 0.10	2.5	2.5	2.2	1.4	2.2
1,500	3.6	4.2	2.4	3.0	5.6	3.1	2.8	2.5	3.8	3.3	3.2	2.4	2.7	4.0	3.2	3.6	2.1	4.0	2.8	3.6	3.29 ± 0.12	2.4	2.5	2.4	2.8	2.5
2,000	2.7	4.4	3.4	2.6	3.0	2.9	3.3	3.3	3.5	2.9	4.0	4.3	2.5	2.6	3.0	3.1	4.1	2.7	3.5	2.5	3.27 ± 0.09	2.2	2.7	2.7	2.7	2.6
2,500	3.4	3.1	4.4	3.4	3.7	1.2	1.6	3.3	3.7	3.2	2.2	3.8	4.7	3.2	3.6	2.2	2.8	2.6	3.5	3.8	3.17 ± 0.13	2.8	1.5	2.8	2.5	2.4
3,000	3.3	3.3	3.7	3.7	1.7	3.5	5.2	2.9	3.4	3.7	5.8	4.6	3.3	3.7	4.2	3.5	2.7	4.0	3.6	3.5	3.66 ± 0.13	2.4	2.7	3.3	2.8	2.8
3,500	3.4	2.6	3.9	2.4	3.7	2.6	3.4	3.4	3.9	3.2	3.8	3.4	3.1	3.1	3.1	3.8	3.9	2.5	2.6	5.1	3.40 ± 0.09	2.1	2.2	2.5	2.8	2.4
4,000	2.0	1.7	3.2	3.1	2.4	3.6	2.8	3.9	2.8	4.3	4.3	3.7	3.7	4.5	4.8	4.1	3.4	2.6	4.1	3.5	3.43 ± 0.12	2.0	3.0	2.5	3.0	2.6
4,500	4.1	2.7	3.4	2.9	3.2	2.7	3.3	4.3	3.6	4.0	2.2	4.3	3.5	3.5	3.5	3.6	4.4	2.3	3.5	4.3	3.47 ± 0.10	2.3	2.7	3.1	2.5	2.7
5,000	2.7	3.0	4.1	5.0	5.8	3.7	6.0	3.5	3.3	4.5	5.0	3.1	4.0	3.4	2.9	3.9	4.7	4.1	4.6	2.3	3.98 ± 0.15	3.4	3.3	2.5	3.0	3.0
5,500	3.0	2.1	2.7	3.7	3.2	2.4	2.4	2.9	1.7	1.6	2.1	3.0	3.4	3.3	4.0	3.3	2.1	3.5	2.4	3.0	2.79 ± 0.09	2.3	1.6	2.8	2.4	2.3
6,000	2.1	1.4	1.8	2.2	2.4	2.2	5.1	4.0	3.3	3.7	3.8	6.1	4.1	4.4	3.2	2.2	2.8	0.8	1.6	2.3	2.98 ± 0.18	1.7	3.0	2.6	1.7	2.3
6,500	3.6	3.0	3.2	3.1	4.6	3.3	2.7	1.5	3.0	1.6	2.4	3.2	2.4	1.1	2.0	3.5	3.5	3.3	3.3	4.6	2.95 ± 0.13	3.0	2.0	2.4	2.4	2.5
7,000	1.5	1.0	1.5	1.9	1.3	3.3	3.6	2.5	3.1	2.3	4.0	2.0	3.8	2.9	2.6	5.2	3.1	3.7	3.1	3.8	2.81 ± 0.15	1.5	2.4	2.0	2.6	2.0
7,500	3.2	5.0	2.2	4.0	2.6	3.0	3.4	3.2	3.9	2.5	2.7	2.1	2.2	2.7	1.8	2.5	2.3	4.1	2.0	2.4	2.89 ± 0.12	2.5	2.4	2.3	2.3	2.4
8,000	1.0	1.5	0.6	1.0	1.0	2.1	1.8	2.4	1.4	2.5	1.9	2.6	2.4	2.9	2.0	2.5	2.5	2.5	2.7	3.1	2.02 ± 0.10	1.1	1.7	2.0	1.3	1.5
8,500	2.6	1.0	0.9	1.5	1.0	1.6	1.2	1.9	1.1	1.9	1.5	2.0	2.0	1.7	1.0	1.2	1.1	2.5	2.7	2.7	1.60 ± 0.09	0.9	1.1	1.6	1.5	1.3
9,000	1.7	1.6	1.6	1.4	0.0	2.0	1.5	0.6	1.9	0.0	1.3	1.2	1.0	1.0	1.7	1.8	1.6	1.4	0.5	1.6	1.40 ± 0.06	1.0	1.0	1.5	1.0	1.2
9,500	1.9	1.2	2.0	1.3	2.0	1.0	0.6	0.9	0.0	0.0	1.5	0.5	0.75	0.9	0.3	..	..	..	..	1.9*	1.20 ± 0.08	1.4	0.7	0.8	1.5	1.4
10,000	0.8	1.8	1.2	1.6	0.6	0.9	0.9	1.1	0.7	0.7	..	..	..	..	0.6*	..	..	..	..	1.9*	1.00 ± 0.07	1.2	0.2	0.2	0.6	0.6

* Total top weight.

concentrations from 500 to 7500 p.p.m. is a very striking fact in itelf. But the extreme resistence of the wheat plant to the effects of NaCl is even more striking and the behavior of the plants in their earlier stages of growth, showing an initial depression in root and top development at the lower concentrations of 500 and 1000 p.p.m. NaCl and a stimulation beyond those points, is perhaps the most striking fact of all and one which is extremely difficult to explain on the basis of our present knowledge.

ANALYSES OF PLANT ASH IN THE WHEAT SERIES

Evidence has been adduced repeatedly in support of the more or less close relationship which exists between the absorption of ions by roots and the quality and quantity of plant growth attained. Knowledge on this point, however, is at the present time fragmentary and does not allow formulation of any quantitative idea with respect to this relationship. Although the task is an extremely laborious one, the authors deemed it worth while to carry out analyses of the ash of plant tops in the foregoing wheat series. The results of these analyses are set forth in table 3. The irregularity of the data given testifies to the errors to which analytical procedures in this regard are subject. There are, nevertheless, some interesting lessons, or at least interesting indications, attaching to the analytical data in question.

The plant seems to possess a mechanism for regulating its total ash content within certain fairly narrow limits, until a high concentration of NaCl is reached, when unusually large absorptions of salts occur which are reflected in the total ash contents of the plants. It is interesting to note that the marked increase in the ash content of these plants, which makes a pronounced deviation from a fairly uniform ash content in all the other series, is found in the plants grown in 8000 p.p.m. NaCl. That is the very point where a marked depression in the yield of roots and tops is evident. When the individual constituents of the ash are considered, there are some which require little or no comment. For example, the PO_4, Fe_2O_3, Al_2O_3, and SiO_2 content is reasonably uniform throughout and is seemingly slightly or not at all affected by the changes in the NaCl content of the solutions in which the plants were grown. This is also true apparently for the potassium, though the analytical errors for this element are larger and the variability seems to be slightly greater as a result. In the case of the other ions, however, there are some very striking changes, as between their content in the ash of plants grown without NaCl and in plants grown with varying quantities of NaCl. Probably the most striking of all these is the case of the calcium ion. It will be noticed in a study of the data on calcium of the plants in the different series that even at 500 p.p.m. NaCl, there is a sharp depression over the control in the amount of calcium in the ash. The plants grown in 500 to 1500 p.p.m. NaCl have only one-third to one-half as much calcium as the plants in the controls which received no NaCl. The ash of plants grown in higher concentrations of NaCl than those named contained even less calcium, which, with one or two excep-

tional figures probably representing analytical errors is fairly uniform through out all of the concentrations of NaCl in question. It is certain that there is only approximately one-fourth as much calcium in the ash of plants grown in concentrations of 2.000 p.p.m. NaCl and above as in the ash of the control plants. Whether this depression in the calcium content is beneficial to the growing cells of the wheat remains to be answered by further study. There are some other lines of evidence which seem to converge on the point that such a benefit may be conferred on the wheat plant by reducing its calcium

TABLE 3

Absorption of ions from solutions containing NaCl as shown by ash analysis of wheat plant tissue

CONCENTRATION NaCl	PERCENTAGES, DRY WEIGHT											
	Total ash	SiO_2	Al_2O_3	Fe_2O_3	Ca	Mg	Cl	SO_4	PO_4	Na	K	N
p.p.m.												
000	7.91	0.15	0.09	0.04	1.11	0.20	0.12	2.31	1.12	0.09	1.73	1.97
500	6.09	0.07	0.06	0.02	0.41	0.21	0.63	1.00	1.23	0.67	1.50	1.15
1,000	6.62	0.09	0.07	0.03	0.42	0.01	0.94	0.95	1.17	0.32	1.75	*
1,500	8.95	0.14	0.07	0.03	0.51	0.20	1.45	1.19	1.11	1.17	1.70	1.16
2,000	7.46	0.14	0.06	0.03	0.37	0.18	1.46	0.92	1.10	1.16	1.44	1.49
2,500	7.15	0.18	0.05	0.04	0.31	0.01	1.44	0.67	1.07	1.24	1.36	*
3,000	6.98	0.09	0.05	0.02	0.26	0.13	1.55	0.69	1.03	1.45	1.50	1.33
3,500	7.80	0.10	0.06	0.02	0.30	0.13	1.80	0.86	1.13	1.30	1.56	1.05
4,000	7.23	0.11	0.06	0.02	0.20	0.09	1.86	0.66	1.02	1.17	1.62	1.33
4,500	8.00	0.22	0.09	0.03	0.25	....	1.79	0.72	0.97	1.50	1.38	1.21
5,000	7.13	0.10	0.09	0.02	0.62	....	1.64	0.60	0.87	1.27	1.59	1.12
5,500	7.43	0.09	0.05	0.02	0.21	0.10	1.92	0.56	1.08	1.12	1.64	1.24
6,000	8.35	0.11	0.06	0.02	0.28	0.12	2.71	0.50	0.94	1.41	1.86	1.29
6,500	6.97	0.09	0.10	0.02	0.14	0.08	1.79	0.04	0.99	1.00	1.14	1.03
7,000	6.93	0.10	0.08	0.02	0.15	0.12	1.88	0.51	0.95	1.19	1.47	1.01
7,500	8.89	0.17	0.06	0.03	0.22	0.01	2.65	0.54	1.00	1.15	1.24	1.15
8,000	10.42	0.09	0.15	0.03	0.24	0.14	3.73	0.47	1.19	2.25	1.71	1.56
8,500	10.11	0.11	0.07	0.02	0.27	0.14	3.62	0.68	1.09	2.14	1.53	1.61
9,000	11.70	0.12	0.06	0.02	0.26	0.11	4.66	0.69	1.38	2.27	1.83	....
9,500	12.65	0.08	0.17	0.02	0.32	0.04	5.14	0.70	1.12	2.62	1.81	....

* Lost.

content. In regard to the magnesium we find a similar situation as that which obtains in the case of the calcium, but the depression is, perhaps, not quite so marked as in the former case. It is noteworthy, however, that in the ash of some of the plants receiving NaCl little or no magnesium is found. This. too, may have a greater physiological significance than we are able, in the light of our present knowledge, to discern. So far as the chloride and sodium ions are concerned, only brief comment is necessary. Obviously with the increase of NaCl in solution an increase in the sodium and chloride of the plant ash is expected. It is noteworthy, however, that over a large range of concentrations of NaCl there is a fairly constant (analytical errors being taken into ac-

count) amount of chloride in the ash of the plants. Here again as in the case of the total ash we find a sharp and rapid rise in chloride content above 7000 p.p.m. NaCl in the culture solution. The same is true for the sodium ion, where the rise is much more sharp and begins at 8000 p.p.m. NaCl. It is interesting to note besides that, as several other recent investigations on absorptions of ions have indicated, the sodium and chloride ions evidently do not enter the plant or reach the tops at the same rates. Moreover, the absorption of chloride ions is noticeably faster at the lower concentrations of NaCl than at the higher concentrations of NaCl. To what extent this phenomenon is related to the permeability of the cells of the root and the top, is an open question. Not the least interesting of all the ions, is the sulfate ion. It will be noted that the introduction of 500 p.p.m. NaCl into the control solutions depresses the amount of the sulfate ion in the ash to a very marked degree. This depression lasts to the same extent up to a concentration of 2000 p.p.m. NaCl, when there is a further marked depression which remains fairly constant for all of the higher concentrations of NaCl. Apparently the chloride ion in some way substitutes itself for the sulfate ion or prevents the latter's entrance at the normal rate. All in all, it would seem that studies on the ash constituents of plants grown in such solutions are, in spite of the labor involved, well worth while, especially if the analytical methods, can be so much farther refined as to reduce the limits of error appreciably.

For a somewhat different reason than the other data, we have also included in table 3, figures on the total nitrogen of the plants grown in all the series. It is perfectly clear that NaCl in all concentrations reduces the percentage of nitrogen in the wheat plant to a marked extent. Even the smallest amount of NaCl used, namely 500 p.p.m., depressed the nitrogen content of the dry matter by approximately 40 per cent. This is probably indicative of the interference by the chloride ion with the penetration of the nitrate ion just as it interferes apparently with the penetration into the cell of the sulfate ion. It is evident from the figures on the nitrogen content of the dry matter in these plants that the total yield and appearance of a plant, as has been shown be-before, may give no indication of the nitrogen content thereof. A limited nitrogen supply at the disposal of the root systems would seem to be adequate for making maximum growths of plants if other conditions are satisfactory.

RESULTS OF THE BARLEY SERIES

The experiments with barley were carried out in the winter and spring of 1921–22. The details of the experiment, which are somewhat different from those of the wheat series, will be described separately.

The nutrient solution employed was the same for the barley series as for the wheat series but the NaCl was added to the several cultures in concentrations which did not comply, as regards intervals between successive concentrations, with the system employed in the wheat series. Each treatment was supplied in sextuplicate instead of quadruplicate as in the wheat series. This allowed

of 30 plants for every treatment and gave a better basis for statistical treatment of the data obtained at harvest. Table 4 gives the data with regard to the concentration of the NaCl used in the different cultures. An accident, through which most of the plants in one of the barley series were burned, made necessary the running of another series in the spring of 1922. The results on the plants saved from the first barley series, which was run in the fall of 1921, are also given in tables 6 and 7, because of some interesting facts which they bring out. Moreover, the water lost by transpiration was determined throughout the barley series, because the authors thought it would be interesting to determine how the salt content of the solutions influenced water losses under the conditions obtaining in this experiment. The methods used

TABLE 4

Tolerance of barley plants for NaCl

Dry weights, water transpired, and derived functions

NaCl	DRY WEIGHT TOPS PER PLANT	P.E. MEAN	DRY WEIGHT ROOTS PER PLANT	TOTAL DRY WEIGHT PER PLANT	H_2O TRANSPIRED BY 30 PLANTS	RATIO OF H_2O TRANSPIRED, TO DRY WEIGHT
	gm.		*gm.*	*gm.*	*liters*	
0	3.30	±0.08	0.48	3.78	68.8	610
500	3.82	±0.09	0.46	4.28	70.7	560
1,000	3.45	±0.11	0.41	3.86	66.3	550
1,500	3.58	±0.11	0.40	3.91	61.9	530
2,000	4.02	±0.10	0.65	4.47	62.0	480
3,000	3.98	±0.14	0.41	4.39	52.2	420
4,000	3.66	±0.13	0.42	4.08	44.0	390
5,000	3.56	±0.14	0.42	3.98	41.6	370
6,000	3.66	±0.13	0.34	4.00	33.2	310
7,000	3.13	±0.19	0.24	3.37	21.8	320
9,000	0.71		0.72	0.83	7.3	310
11,000	0.46		....	....	3.2	260
13,000	0.20		....	....	1.1	...
15,000	0.11		....	....	....	...

in germinating the seeds and in setting out the plants were the same as those used in the wheat series. The solutions were changed once during the growing period. The second series of barley plants having been set out on January 26 and harvested on April 11, the change of solution took place on March 20. It was observed from the beginning that barley plants may continue to live for some time in high concentrations of NaCl without making any evident growth. In fact, the leaves of the plants may be dead and the roots look unhealthy, and yet the plants may be transferred to solutions free from NaCl and they will begin to make growth and continue to grow normally.

Seven days after placing the plants in the solutions, those in concentrations of 18,000 p.p.m. NaCl or in excess thereof had turned yellow and were making no growth. Plants in concentrations of 21,000 p.p.m. NaCl finally died with-

out making any growth, Plants in concentrations of 15,000 p.p.m. NaCl and below showed growth by actual elongation of the stem and the formation of new leaves. The difference between the behavior of the plants in concentrations of 15,000 to 18,000 p.p.m. NaCl is quite distinct. Table 5 indicates the number of plants which died in the higher concentrations and the time after planting at which such deaths took place. The death of many of these plants occurred only after the plant had made considerable growth. This emphasizes again the point which has been made in other papers issued from this laboratory, that as nearly as possible the full growing period of the plant must be allowed in such experiments. Table 6 indicates similar results

TABLE 5

Tolerance of barley plants for NaCl

The number of plants in the various concentrations dying before completion of experiment*

TIME FROM PLANTING	CONCENTRATION OF SOLUTION IN P.P.M. NaCl						
	6,000	7,000	9,000	11,000	13,000	15,000	18,000
days							
10–							
15–							
20–			1	1	2	4	7
25–							
30–		1			1		
35–				4	7	6	
40–							21
45–	1		1	2	3	3	
50–		1	3		4	1	
55							
60–							
65–		3	5	10	3	7	
70–							
75–			12	4	9	5	1
80–							
Total....	1	5	22	21	29	26	29

* Thirty seedlings originally in each solution.

obtained with the plants in the first barley series. At the time of harvesting, all the plants in concentrations of 9,000 p.p.m. NaCl and below were generally in a healthy condition. It will be seen from a study of table 4, in which the dry weights of the tops and roots of the individual plants are given, that the dry weight of barley plants was greater in all the concentrations of NaCl from 500 to 6,000 p.p.m. inclusive, than in the control series which received no NaCl. The appearances of the plants were in accord with the dry weighs, inasmuch as the heights of all the plants receiving NaCl up to concentrations of 6000 p.p.m. were greater than the heights of the plants receiving no NaCl. It will also be noted in table 4 that there is a very definite and marked de-

pression in the water requirement of the barley plants after the NaCl content of the culture solution is increased. As a coincidence, it is noticed that here, as in the wheat series, the concentration of 8000 p.p.m. NaCl seems to be the critical one where the marked depression in plant growth occurs.

The two maxima for roots and tops which were noted in the case of the wheat series are not apparent in the barley series, and the stimulating effect of NaCl to barley plants under the conditions of the experiment is not nearly so marked as in the case of the wheat series. The roots were much more de-

TABLE 6

Tolerance of barley plants for NaCl

The number of plants in the various concentrations dying before completion of experiment*

TIME FROM PLANTING	CONCENTRATION OF SOLUTION IN P.P.M. NaCl															
	7,500	8,000	8,500	9,000	9,500	10,000	10,500	11,000	11,500	12,000	12,500	13,000	13,500	14,000	14,500	15,000
days																
10–																
15–																
20–																
25–																
30–														1		
35–						1					2					
40–																
45–																
50–																
55–				3	1	5	6	9	6	11	8	5	8	8	9	9
60–																
65–																
70–				1	5		8	5	10	14	14	15	15	13	13	10
75–																
80–																
85–																
90–	2	3	13	11	16	8	3	2			5		7	7	7	10
Total..	2	3	13	15	22	14	17	16	16	25	29	20	30	29	29	29

* Thirty seedlings originally in each solution.

pressed in their development at concentrations beyond 5000 p.p.m. NaCl than were the tops, the differences between the 5000 and 6000 p.p.m. concentrations being very marked. In concentrations of 13,000 p.p.m. NaCl and above, the roots made practically no increase in length and were markedly discolored. In consonance with the results obtained by Muenscher (1) and others, the authors' data show that there is no parallelism between the amount of water transpired and the dry weights of barley plants produced. The curve shown in figure 1 obtained for the dry weights produced and the water transpired (curve B), indicates that there is a continuous fall in the curve

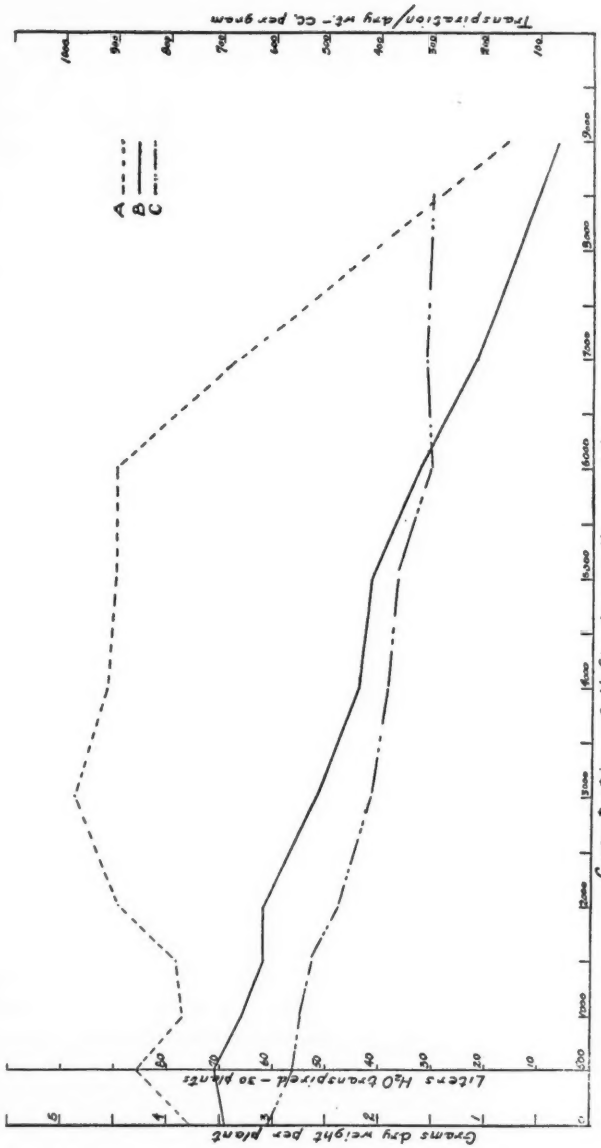

FIG. 1. THE EFFECT OF VARIOUS CONCENTRATIONS OF NaCl ON THE GROWTH OF BARLEY
A = total dry weight; *B* = total water transpired; *C* = ratio of water transpired to total dry weight

for water transpiration but an appreciable rise in the curve for the dry weight (curve A), and a fall when a sufficiently high concentration of NaCl is reached. This same figure gives an interesting comparison between the transpiration coefficient and the concentration of the medium (curve C). It will be seen that until concentrations of over 6000 p.p.m. NaCl are reached, the points fall very close to a line which is practically straight. A comment will be made later on the probable cause of the difference in the response between wheat plants and barley plants to the several concentrations of NaCl.

We have referred above to some interesting results which were obtained from the first series of barley with those plants in the series which were not burnt. Table 7 gives the results in question. It will be noted in that table that 500 p.p.m. NaCl proved to be markedly stimulating to the barley plants but that no other concentration of NaCl among those recorded in table 7 gave stimulation to barley growth over that obtained in the control. In fact, in the solutions containing 2000 p.p.m. NaCl, there seems to be a slight depres-

TABLE 7

Tolerance of barley plants for NaCl

Dry weights, water transpired, and derived functions—first experiment

NaCl	DRY WEIGHT TOPS PER JAR	P.E. MEAN	DRY WEIGHT ROOTS PER JAR	P.E. MEAN	TOTAL DRY WEIGHTS PER JAR	RATIO $\frac{\text{TOP}}{\text{ROOTS}}$	P.E. MEAN
p.p.m.	*gm.*		*gm.*		*gm.*		
0	7.94	±0.28	0.89	±0.049	8.85	9.0	±0.37
500	12.57	±0.69	2.16	±0.140	14.73	5.7	±0.17
1,000	7.38	±0.05	1.06	±0.057	8.44	6.9	±0.17
1,500	7.59	±0.27	0.87	±0.062	8.46	8.6	±0.50
2,000	6.36	±0.05	0.99	±0.120	7.35	7.0	

sion in the growth of barley. It is interesting to compare these results with those obtained in the second barley series which are given in detail above. It seems obvious that the climatic complex under which plants are grown in such experimental work is of great moment in determining the effect of other environmental factors on plant growth. The very marked stimulation to barley growth exerted by 500 p.p.m. NaCl in this case is very striking. It is noteworthy, besides, that the ratio of tops to roots in regard to the dry matter produced is much higher in the control than it is in the cultures which received NaCl, and peculiarly enough, where the stimulation of the barley plants is obtained, the ratio was the lowest. The reason for this is that the roots in the cultures containing 500 p.p.m. NaCl had been stimulated so much more than the tops as to lower the ratio. The marked difference in the degree of stimulation of roots and tops in the cultures receiving 500 p.p.m. NaCl held throughout the growing period of the plants and was noticeable a few days after the plants were set out.

RESULTS OF THE PEA SERIES

Only three seedlings per jar were set out in the case of the pea series. Otherwise, the same culture solution was used as in the other series. The concentrations of NaCl employed and the data obtained are given in table 8. The experiment was started January 28, 1922, and the plants were harvested on March 28. Dwarf garden peas were used. The weather was rather cold during the early part of the growing period of these plants but conditions improved in the latter part of the growth period. Nodules were noted on the plants in only a few jars in the large series which was arranged in this experiment. The development of the nodules did not seem to have any effect on the reaction of the plants to the NaCl. Flowers were observed on February

TABLE 8

Tolerance of pea plants for NaCl

Dry weights, water transpired, and derived functions

NaCl*	DRY WEIGHT TOPS PER JAR	P.E. MEAN	DRY WEIGHT ROOTS PER JAR	TOTAL DRY WEIGHTS PER JAR	WATER TRANSPIRED, 6 JARS	RATIO WATER TRANSPIRED, TO DRY WEIGHT
p.p.m.	*gm.*		*gm.*	*gm.*	*liters*	
0	6.48	±0.18	0.91	7.39	16.1	370
500	6.84	±0.16	0.96	7.80	16.9	360
1,000	6.88	±0.34	0.99	7.87	16.6	360
1,500	7.32	±0.23	0.91	8.23	15.6	320
2,000	6.18	±0.17	0.94	7.12	14.1	330
3,000	6.24	±0.06	1.07	7.31	12.4	310
4,000	5.78	±0.23	0.84	6.62	10.3	260
5,000	4.46	±0.20	0.86	5.32	8.5	270
6,000	2.89	±0.23	0.65	3.54	6.2	230
7,000	1.92	±0.20	0.47	2.39	2.9	230
9,000	0.73	±0.03	0.01	0.74	0.8	...

* In addition to the concentrations of NaCl here noted, the following were employed: 11,000, 13,000, 15,000, 18,000, and 20,000. For observations and remarks, see test.

23 and when the plants were harvested, many of them bore well-filled pods. Flowering continued until the plants were harvested. No correlation could be found between different concentrations of NaCl used and the time of flowering, flowers being produced irregularly throughout the series from the beginning of the flowering period to harvesting time. The effect of NaCl on the roots of the pea plants in the jars containing the higher concentrations of NaCl was obvious from the beginning of the experiment. Five days after setting out the plants it was noticed that in the solution containing 11,000 p.p.m. NaCl, the plants had produced very few lateral roots and in the solution containing 13,000 p.p.m., practically no lateral roots were produced. The plants in the solution containing 15,000 p.p.m. NaCl had produced no new roots, although the tops were growing. Later it was noticed that in

solutions containing 6000 p.p.m. NaCl and above, the roots were swollen in a peculiar manner. The condition of the roots in solutions of 11,000, 13,000, and 15,000 p.p.m. NaCl remained thus until the death of the plants. The swollen condition of the roots goes so far as to manifest itself in an apparent bursting of the tissue so as to show longitudinal cracks in the roots. Other investigators have referred to this peculiar condition of plant roots grown in solutions of high osmotic pressure. The seedlings in solutions of 20,000 p.p.m. NaCl and above died within a few days. Two weeks after planting, the seedlings in solutions containing 18,000 p.p.m. NaCl and above were dead, not having made any growth, whereas with few exceptions, seedlings in concentrations of 15,000 p.p.m. NaCl and below were green, and had made visi-

TABLE 9

Tolerance of pea plants for NaCl

The number of plants in the various concentrations dying before completion of experiment

TIME FROM PLANTING	CONCENTRATION IN P.P.M. NaCl						
	5000	6000*	7000*	9000	11000	13000	15000
days							
10–							
15–							
20–							
25–						8	6
30–							
35–				2	7	6	10
40–	1	1	2	3			
45							
50–		5	1	6			
55–			2				
60–				All plants dead			

Eighteen seedlings originally in each solution.

* In these two concentrations 3 of the plants recorded in each were in one jar. Their death was evidently due to a factor common to the jar.

ble growth. The difference in behavior between the seedlings in concentrations of 15,000 and 18,000 p.p.m. NaCl was distinct throughout the two sets and showed a critical point above which plants could not survive under these experimental conditions, but below which growth was possible. Plants placed in some concentrations below this critical point died after making a certain amount of growth. Table 9 shows the number of plants that did not survive the various concentrations up to the time of harvesting. Those data indicate that the plants in concentrations of 11,000 p.p.m. NaCl and above died within forty days after planting. The stems of these plants increased in length and the number of leaves increased, but the change in weight was insignificant. Unlike those of the barley, the leaves of these plants withered from the edges inward just before the plants were harvested. All

plants in concentrations of 9000 p.p.m. NaCl died at approximately the same time throughout the jars of that concentration. This emphasizes again the point which we stressed in connection with the barley and the wheat series, namely, that no one part of a plant's growth period constitutes a reliable criterion for any environmental effect. Before the plants in 9000 p.p.m. NaCl died, their tops had grown 5 inches and the roots 7 inches. The plant had flowered and small pods about one inch in length had been produced.

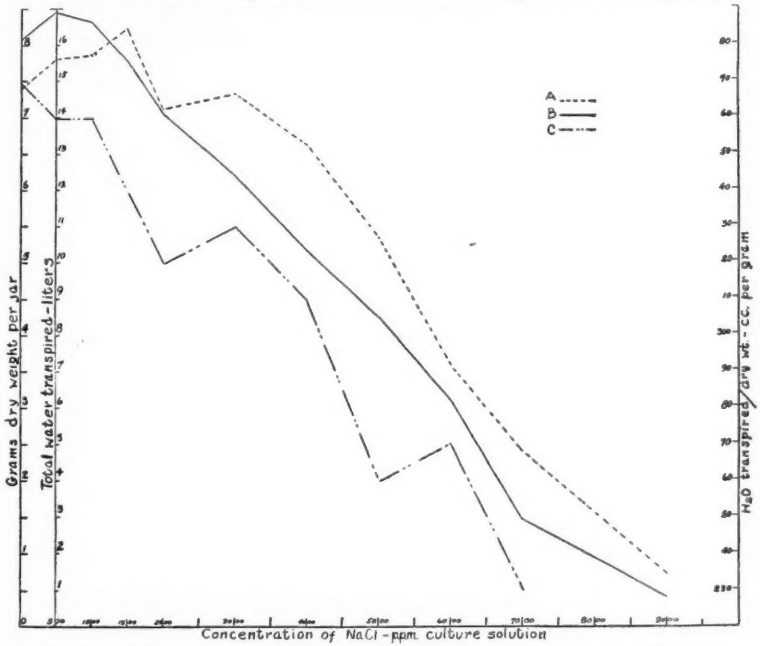

FIG. 2. THE EFFECT OF VARIOUS CONCENTRATIONS OF NaCl ON THE GROWTH OF GARDEN PEAS

A = total dry weight; B = total water transpired; C = ratio of water transpired to dry weight.

The deaths of a number of plants at lower concentrations may safely be ascribed to individual variations.

Table 8 also sets forth the results obtained in the pea series on dry weights of tops and roots, and indicates the water transpired by the plants. The relationships between the water transpired and the dry weights obtained are plotted in figure 2. Because of the great individual variability of pea plants, which is characteristic as far as is known, the probable errors of the results are high. It will be seen, nevertheless, that up to concentrations of 4000 p.p.m. NaCl, there are no significant differences in the dry weights among the

several cultures. Obviously, the differences might have been shown to be greater if the variability had not been so great. No definite relation seems to have subsisted between the tops and the roots produced in the concentrations of NaCl employed in the cultures. The curve for the water transpired follows in general the curve for total weight, but the correlation holds only within certain limits, since the former curve falls off more rapidly than the latter. In figure 2 (curve C) the transpiration coefficient is plotted against the concentrations of NaCl employed. It is seen from this that within the large area involved, the coördinates lie near a straight line.

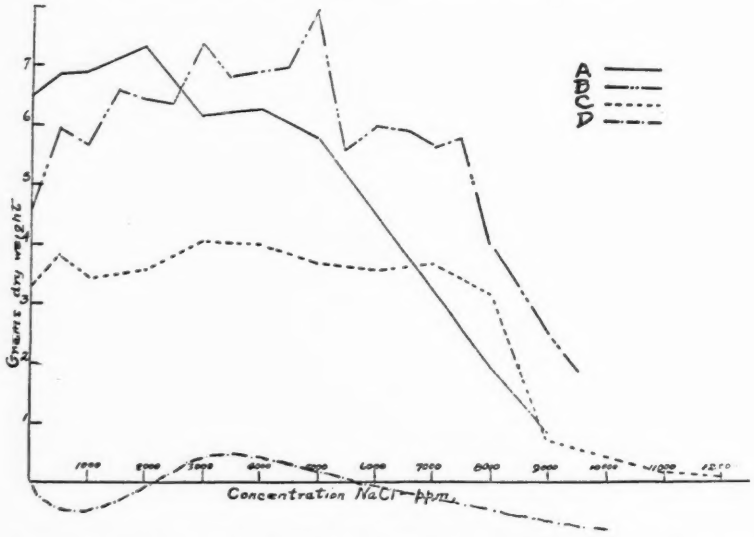

FIG. 3. A COMPARISON OF THE EFFECT OF VARIOUS CONCENTRATIONS OF NaCl ON THE GROWTH OF WHEAT, BARLEY AND PEAS

A = peas; *B* = wheat; *C* = barley, *D* = relative length of wheat roots after 3 weeks (diagrammatic).

As has already been indicated, the variability which characterizes pea plants does not permit us to say definitely whether concentrations of NaCl up to and including 3000 p.p.m. are stimulating to pea plants. There would seem to be some slight evidence that up to and including concentrations of 1500 p.p.m. NaCl, a slight stimulation does, nevertheless, exist. It is perfectly clear, on the other hand, that irrespective of the variability, the point of incipient toxicity of NaCl is marked with a fair degree of clearness. It appears to be at a concentration of 4000 p.p.m. NaCl. Increases of salt above that figure rapidly decreased to a marked degree the growth of the pea plants until at a concentration of 9000 p.p.m. NaCl, very little dry weight

of pea plants was produced. This also indicates that the pea plant is much more susceptible to the toxic effects of NaCl than barley or wheat plants, inasmuch as the toxicity sets in at lower concentrations of the salt. In fact, it would seem from results obtained from these series conducted under approximately the same environmental condition, that the point of definite toxicity of NaCl for pea plants is at a concentration of only slightly more than half that for barley or wheat. A graphic comparison of curves obtained for wheat, barley, and peas, is shown in figure 3. The relative length of roots in the wheat series is given by a diagrammatic curve based on observations taken at the end of the first 6 weeks of growth. Other features of the data in tables 8 and 9 require no comment.

GENERAL DISCUSSION

In all the experiments which have been described in this paper, including the wheat, the barley, and the pea, the toxic effects of NaCl, when they set in, manifested themselves with a fair degree of sharpness. For example, none of the plants grew to any appreciable extent above a concentration of 15,000 p.p.m. NaCl. On the other hand, with very few exceptions, growth did take place to some slight extent in a concentration of 15,000 p.p.m. NaCl. Our results, moreover, render it very clear that under controlled experimental conditions in which soil factors, including antagonism between ions, are eliminated from consideration, grasses may be stimulated by large concentrations of NaCl and may be unaffected by even larger concentrations. Although the peas do not manifest so high resistance and probably little or no stimulation from the effects of NaCl, they do nevertheless, exhibit a resistance to fairly large quantities (about 3000 p.p.m.). As has been noted, with one and the same plant in an experiment carried on at different parts of the year, results might show variations. This striking situation was brought home to the authors in subsequent experiments, which are not described in this paper, in which barley and wheat were grown outdoors during the summer season and in which none of the stimulating effects which are described above were noted, even though the resistance to the effects of high concentrations of NaCl was similar to that noted in the experiments described in this paper. The nature of the climatic complex in the environment would, therefore, seem to play a rôle of the greatest importance in determining the effectiveness of another environmental factor, as in the case of NaCl in the root medium. It is not difficult to guess at a possible explanation for this difference in the effectiveness of an environmental factor on a given plant under different climatic complexes. For example, any temperature-humidity-light complex which is favorable to a high transpiration would obviously improve the chances for a large absorption of NaCl. This would result in a marked change in the qualitative nature and concentration of the cell sap, and one would, therefore, expect that both photosynthesis and the normal metabolism of the cells would be markedly influenced thereby in the direction of depressing the

total weight. On the other hand, with a temperature-humidity-light complex in which transpiration was relatively low, the absorption of NaCl by the cells would be relatively low, and hence very different conditions would regulate photosynthesis and metabolism in the cells. Whether this one of the several possible explanations for the facts which are noted is correct, it is emphatically true that the climatic complex is an important determinant of the results that can be obtained with the same plant in experiments in which other factors are perfectly controlled. To show how striking the discrepancy between different sets of results on the same plant may be, one of the authors will publish later the results obtained by him with wheat and barley plants grown at a different season of the year but otherwise under similar conditions to those used above. However that may be, some of the higher green plants seem to possess a great resistance to NaCl and the effects which are so frequently found in alkali soils with relatively small concentrations of NaCl must evidently be ascribed not to the direct effects of NaCl but to some indirect effects exerted by it on the physico-chemical condition of the soil. The hydrogen-ion concentration of the culture solution was apparently a factor of no significance in connection with the results obtained, inasmuch as it varied fairly uniformly throughout the series, and, although started at a point approximately pH 4.9, always reached a pH of 6.7 in a relatively short time and maintained that concentration throughout the growth period of the plants.

SUMMARY

1. A study was made in solution cultures on the effects of a wide range of concentrations of NaCl on the growth of wheat, barley, and peas. Cultures were sufficiently replicated to control errors due to variability.

2. All plants tested show a very high resistance to NaCl.

3. Under some environmental conditions, NaCl is highly stimulating to wheat, even at concentrations of 4000 p.p.m. or more.

4. Small concentrations of about 500 to 1000 p.p.m. NaCl may depress growth particularly in the early stages, but higher concentrations may stimulate it.

5. All plants tested may make growth, though it be arrested with very high concentrations of NaCl, even up to 10,000 p.p.m. or more.

6. The concentration of NaCl at which most marked depression occurs is about 8000 p.p.m.

7. Environmental conditions are a very important determinant of the kind of results which can be obtained.

8. Some of the most striking features of these investigations can be noted only by a careful study of the whole paper.

REFERENCE

(1) MUENSCHER, W. C. 1922. Effect of transpiration on absorption of salt by plants. *Amer. Jour. Bot.* 9: 311–329.

THE ORIGIN AND NATURE OF THE SOIL ORGANIC MATTER OR SOIL "HUMUS": III. THE NATURE OF THE SUBSTANCES CONTRIBUTING TO THE FORMATION OF HUMUS[1]

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The great majority of investigations dealing with the subject of transformation of organic matter in the soil have been concerned either (*a*) with the decomposition of a single purified material, whereby one or more of the intermediary or final products of the reaction were measured, or (*b*) with the change into "humus" of some complex natural organic substance such as cereal straw, corn stover, legume hay, or wood products.

The origin of this so-called "humus," or alkali-soluble organic matter, in the soil is still the subject of various hypotheses, most of which are based upon few experimental facts. On the one hand "humus" was believed to be formed in the soil as a result of "humification" processes carried on by various groups of microörganisms; the nature of the organisms and the chemistry of the processes being very vague. On the other hand, some investigators (6, 7) found that the organic matter added to the soil already contains alkali-soluble material, which actually decreases quantitatively in the soil as a result of decomposition. These investigators claim that there is no specific "humification" in the soil, that the "humus" is actually added to the soil, that it is not formed there, and that it is even gradually decomposed in the soil.

With the exception of a few final products, such as ammonia accumulated and carbon dioxide evolved, very little is known concerning the numerous changes whereby rapidly decomposing natural organic substances are transformed, finally yielding the more or less resistant "humus" materials. This is due largely to the fact that, in the great majority of investigations, a single process has been studied, such as ammonia formation or carbon dioxide evolution, sometimes carried on by a single organism, when the latter was considered at all. Often the interest centered entirely upon the fate of a single elemental constituent of the organic matter, usually nitrogen. The various terms of "decay," "humification," "putrefaction" do not stand for any definite chemical or biological processes but merely for certain sets of condi-

[1] Paper No. 278 of the Journal Series. New Jersey Agricultural Experiment Stations, Department of Soil Chemistry and Bacteriology.

[2] The author is indebted to Mr. R. Dubos of this laboratory for assistance in the determinations of the cellulose and lignin.

tions leading to the formation of certain complex preparations. They are, therefore, valueless in attempting to present a scientific interpretation of the chemical or biological transformation of organic matter in the soil, which results in the formation of dark colored substances referred to collectively as "humus."

It is essential to know the fate of every ingredient of the organic matter added to the soil, the various chemical processes through which it changes, and to what extent it contributes to the soil organic matter. After the information concerning the transformation in the soil of the various carbohydrates, proteins, fats, and other constituents of the natural organic matter is available, an attempt should be made to learn how the presence of one substance influences the decomposition of another. A study of the decomposition of the natural organic matter itself could then be undertaken. Only when the processes thus involved are understood, when the various intermediary, final, and transformation products are known, can one expect to learn the nature of the soil organic matter. It has been shown, for example (8), that in the decomposition of celluloses by filamentous fungi, no residual substances are left which could be classified under "humus." About 60 to 65 per cent of the carbon of the celluloses decomposed was liberated as CO_2 and about 30 per cent of the carbon was reassimilated by the organisms active in the decomposition of the celluloses, in the synthesis of their protoplasm. It was suggested that this synthesized mass of organic matter contributes to the soil "humus." Celluloses are thus found not to play any direct part in the formation of soil "humus." Indirectly, however, they, as well as other carbon compounds offering available sources of energy, contribute decidedly to the soil "humus." The decomposition of celluloses under anaerobic conditions is carried out entirely by bacteria, with the formation of various organic acids and the synthesis of a much smaller amount of protoplasm (12). Under aerobic conditions, celluloses may be decomposed by bacteria with the formation of hemicelluloses, pigments (of the carotin group), and bacterial cells (9). The rôle of celluloses in the formation of "humus" thus depends also upon the environmental conditions under which the decomposition takes place. A knowledge of the chemical composition of the organisms concerned in the decomposition of the organic substances, of the mechanism whereby they obtain their energy, of the efficiency of utilization of the available energy, of the nature of assimilation of the nitrogen and minerals necessary for the synthesis of their cells, and of the relation between the available energy and nitrogen transformation, is, therefore, essential. It has been shown, for example (5, 11), that the formation of ammonia from pure nitrogenous substances by pure or mixed cultures of microörganisms depends not only upon the nitrogen content of the substance in question but also upon the ratio between the available energy (carbon) and the nitrogen content. Substances with a narrow C:N ratio gave much more ammonia than substances with a wide C:N ratio.

When fresh organic matter, in the form of straw, green manure, various plant and animal products, or plant stubble is added to the soil, decomposition sets in rapidly. The total rate of the processes of decomposition can be most conveniently measured by the rate of evolution of CO_2. If the organic material is rich in proteins, the rate of formation of ammonia or the accumulation of ammonia and nitrate nitrogen may also be used in following the rate of decomposition. Not all the organic matter is decomposed in the soil, since both the CO_2 and the final nitrogenous substances (ammonia and nitrate) account for only a part of the original material added. A definite part of the organic matter is reassimilated by the microörganisms active in the processes of decomposition. A part of the original organic matter may be left in the form of various intermediary products such as organic acids and higher alcohols, especially under anaerobic conditions, but this part will tend not to persist in the soil and will be further decomposed in the presence of sufficient base.

TABLE 1

Composition of some natural organic materials on the basis of water-free matter (after Pringsheim)

SUBSTANCE	ASH	FATS AND WAXES	CRUDE PROTEIN	PENTOSANS	CELLULOSE PURE	LIGNIN (WILLSTÄTTER METHOD)
	per cent	*per cent*	*per cent*	*per cent*	*per cent*	*per cent*
Winter straw	4.33	0.67	3.00	21.67	34.27	21.21
Summer straw (oats)	4.81	2.02	4.70	21.33	35.43	20.40
Corn stover	6.15	0.77	3.50	23.54	30.56	15.13
Corn cobs	1.80	1.37	2.11	31.50	37.66	14.70
Hay	6.05	2.00	9.31	13.52	28.50	28.25
Reeds	5.79	0.91	3.76	20.94	33.35	20.33
Pine wood	0.53	3.17	1.27	10.80	41.93	29.52

There is another part of the natural organic matter left in the soil, namely, a part of the original material which resists decomposition. It is this undecomposed organic matter, the various intermediary products, and the newly synthesized materials which go to make up the soil organic matter or "humus."

To understand the nature of this "humus," it is essential to know the chemical nature of the various substances which go to make up the original organic matter added to the soil, the processes which each of the chemical constituents undergoes in the soil, the environmental conditions, and the nature of the microörganisms bringing these processes about. Since the organic materials added to the soil are very complex and consist of as many chemical substances as only the complex protoplasm of the different cells of plants, animals, and microörganisms can be expected to contain, and in view of the fact that the soil harbors a great many types of microörganisms varying in their metabolisms, many of which contribute to the transformation of the soil organic matter, one can readily understand that the problem under consideration is not very simple.

Of the different ingredients of the natural organic matter of plant origin commonly added to the soil, it is sufficient to consider, for the present, the transformation of the celluloses, starches, pentosans, and other polysaccharides and monosaccharides, which make up about 40 to 70 per cent of the dry matter; proteins and protein derivatives (1 to 20 per cent); the lignins (10 to (30 per cent); and the ether- and alcohol-soluble substances or fats and waxes (0.5 to 4 per cent), as shown in table 1.

Various natural organic materials also contain soluble sugars, Collison (3) having reported 1.56 per cent reducing sugar in wheat straw, 1.38 per cent in corn stover, 5.97 per cent in timothy hay, and 4.35 per cent in alfalfa. The amount of sugars as well as of starches and pectins depends not only upon the nature of the plant but also upon the degree of its maturity.

The problem of the origin of "humus" in the soil thus reduces itself to the following questions:

1. To what extent do the natural organic substances contribute directly, in the form of various ingredients, to the soil "humus"? 2. What are the chemical and biological processes whereby the natural organic substances are decomposed, and how do these contribute to the soil "humus"? 3. What rôle do microörganisms play in the transformation of energy in the soil and how much of this energy is stored away in newly synthesized protoplasm? What relation does this synthesized protoplasm have to the soil "humus"? 4. How do the activities of the microörganisms influence the composition of the "humus," especially its carbon and nitrogen relationship?

EXPERIMENTAL

To determine what constituents of natural organic matter are first decomposed by microörganisms and what constituents resist decomposition, a series of experiments was undertaken on natural organic matter, from which one or more fractions had been removed. The methods outlined by Dore (4), Johnsen and Hovey (10) and others, on wood analysis, were utilized both for removing various ingredients of certain natural products, such as straw, and for measuring the amount of decomposition of each of these ingredients. Cellulose in pure form was determined by the method of Charpentier (2), and cellulose in the form of natural materials was determined by the method of Bengtsson (1). Lignin was determined by the method of Willstätter (14). The alkali-soluble portion of natural organic materials precipitated with hydrochloric acid is often considered as lignin. This preparation, however, contains a considerable amount of pentosans (xylans), which may or may not be utilized as sources of energy by different organisms. These pentosans can be removed, if the precipitate is boiled in the presence of an excess of 2 to 3 per cent acid. Ammonia was determined by leaching the soil with normal KCl solution, then washing with water and distilling the extract with some heavy MgO into standard acid solution.

A series of 10-gm. portions of a uniform quantity of finely ground rye straw was subjected to a series of treatments, at every step two portions of the

treated straw being left for the study of its decomposition by microörganisms. The results of the analyses of this straw by the methods adapted are given in table 2.

An attempt was first made to study the decomposition of various straw preparations by pure cultures of soil organisms. To select a proper organism which would be capable of attacking vigorously the various constituents of the straw, the following preliminary experiments were carried out:

In 300-cc. round bottom flasks, 100-gm. portions of washed white sand were placed; 3-gm. portions of the straw preparations and 25 cc. of a nutrient solution, containing 10 gm. $(NH_4)_2HPO_4$, 2 gm. KCl, 2 gm. $MgSO_4$, and 0.02 gm. $FeSO_4$ in 1000 cc. of distilled water were added to each flask, which was then plugged with cotton and sterilized, at 15 pounds pressure for 1 hour. The flasks were inoculated in duplicate and connected with a respiration apparatus (13), which was kept in the thermostat at 28°C. The following organisms were tested: *Bac. cereus*, a cellulose-decomposing bacterium No. 7, *Zygorhynchus molleri* and a green *Trichoderma*. The carbon dioxide produced in the decomposition of the

TABLE 2

Composition of rye straw

	per cent
Moisture	6.37
Loss on extraction with benzene and alcohol	3.23
Loss on extraction with cold water for 24 hours	7.01
Loss on extraction with 5 per cent NaOH for 24 hours in cold	29.44
(NaOH extract precipitated with HCl, but not boiled in excess of acid	22.33)
(Lignin obtained directly by Willstätter method	18.79)
Loss on extraction with 2 per cent sulfuric acid by boiling 2 hours	21.15
Residual material (largely cellulose)	32.36
Total	99.56

straw was absorbed in standard barium hydroxide solution, which was titrated back, as often as necessary, with standard oxalic acid solution.

At the end of 21 days incubation, the flasks were disconnected and the residual ammonia, residual organic matter, and cellulose were determined in aliquot portions. The residual organic matter was obtained by washing off all the organic matter from the sand upon a eighed filter paper, then drying the paper and contents to constant weight, igniting, and weighing again. The loss in weight indicates the total amount of organic matter left, free from moisture, of the water-soluble fraction and of ash. (Table 3.)

The results indicate quite definitely that Trichoderma and the Bacterium No. 7 are organisms very active in the decomposition of organic matter. The fungus decomposed more of the cellulose as well as more of the total organic matter than the bacterium. It is interesting to note, however, that the latter produced more carbon dioxide than the fungus. This is no doubt due to the fact that the Trichoderma assimilated more of the carbon for the synthesis of its more extensive protoplasm than the bacterium. This can be seen readily from the amount of residual ammonia: although the bacterium liberated 211.95

mgm. of carbon as carbon dioxide and decomposed at least 708 mgm. of water-insoluble organic matter, it assimilated only 17.0 mgm. of ammonia-nitrogen, showing a ratio of 12.5 between the carbon given off as carbon dioxide and the nitrogen assimilated. The fungus liberated only 188.71 mgm. of carbon as carbon dioxide but it decomposed 1228 mgm. of water-insoluble organic matter and assimilated 25.5 mgm. of ammonia-nitrogen, with a ratio of 7.5 between the carbon of carbon dioxide liberated and the nitrogen assimilated. The ratio between the water-insoluble organic matter decomposed and the ammonia-nitrogen assimilated is about the same in both cases, the difference being largely in the amount of carbon liberated as a waste product. This indicates that the bacterium consumed a considerably larger amount of carbon in proportion to the amount of nitrogen assimilated than did the fungus. The fact that bacterial cells are usually richer in nitrogen than the fungus mycelium tends further to emphasize the greater consumption of energy per unit of quantity of cell material synthesized in the case of bacteria than in the

TABLE 3

Decomposition of straw by different microörganisms in sand media

NAME OF ORGANISM	CO_2 PRODUCED, AS CARBON									RESIDUAL AMMONIA (N)	RESIDUAL ORGANIC MATTER*	RESIDUAL CELLULOSE
	2 days	4 days	5 days	8 days	10 days	12 days	14 days	21 days	Total			
	mgm.	*mgm.*	*mgm.*	*mgm.*	*mgm.*	*mgm.*	*mgm.*	*mgm.*	*mgm.*	*mgm.*	*mgm.*	*mgm.*
Control.....	1.97			2.11				5.76	9.84	45.8	2,336	1,020
Bac. cereus.....	2.59	19.15		9.50				26.21	57.45	44.9	2,308	1,016
Bacterium No. 7	2.30		12.96	43.20	27.79	27.36	26.92	71.42	211.95	28.8	1,628	778
Zygorhynchus...	20.45	8.92	10.66	8.35				30.10	78.48	45.8	2,108	1,024
Trichoderma....	21.17	51.64	25.34	39.31	13.10	8.78	12.81	16.56	188.71	20.3	1,380	622

* Free from moisture, water-soluble substances, and ash.

case of fungi. There is ample evidence in the literature to indicate that fungi are much more efficient than bacteria in the utilization of energy. The assimilation of 25.5 mgm. nitrogen by the fungus indicates that at least about 500 mgm. of mycelium has been synthesized, assuming that the nitrogen content of the mycelium is about 5 per cent. This considerable quantity of synthesized mycelium (except the water-soluble portion) is included in the residual organic matter. The fact that fungi take an active part in the decomposition of celluloses in the soil (12), and the fact that they may reassimilate 30 parts of the carbon of the celluloses and other energy sources decomposed, point to the development of these organisms as an important source of "humus" in the soil.

The Zygorhynchus did not attack the celluloses; its activities seemed to have been limited to the soluble carbohydrates and proteins of the straw, as indicated by the fact that it did not assimilate any of the ammoniacal nitrogen. The same is true of the *Bac. cereus*.

To obtain further information concerning the ability of different organisms to attack straw and certain of its constituents, the above experiment was repeated, only 5 gm. of $(NH_4)_2HPO_4$ being used per liter of medium and 25 cc. of the nutrient solution being added to 100-gm. portions of washed sand containing 3 gm. of rye straw. The cultures were sterilized, inoculated with pure cultures of the different organisms, and incubated for 30 days. The determinations were then made as in the previous experiment, and the results are recorded in table 4.

The results show definitely that Trichoderma is most active in decomposing the straw as a whole, nearly 800 mgm. of the water-insoluble, ash-free organic matter being decomposed in 30 days. This is accompanied by a considerable absorption of ammonia nitrogen, which has become transformed into microbial protoplasm. The Zygorhynchus consumed a much smaller amount of the organic matter, accompanied also by a very low ammonia assimilation. The

TABLE 4

*Decomposition of straw by pure cultures of organisms.**

ORGANISM	AMMONIA (N)		RESIDUAL ORGANIC MATTER LEFT	
	Left in 100 gm. of sand medium	Used up by organism	Total organic matter, free from ash and water-soluble constituents	Precipitate formed by treating NaOH solution with excess of acid
	mgm.	*mgm.*	*mgm.*	*mgm.*
Trichoderma	8.4	16.0	1,932	324
Zygorhynchus	22.1	2.3	2,388	490
Actinomyces viridochromogenus	21.4	3.0	2,193	286
Bacillus cereus	19.4	5.0	2,657	496
Control	24.4	0	2,729	504

* Three grams of straw, containing about 2700 mgm. of dry matter free from ash and water-soluble constituents, added to 100 gm. of white, washed sand.

Bac. cereus decomposed only a small amount of the straw, probably only the reducing sugars and certain readily soluble substances. The actinomyces decomposed a considerable amount of the straw (536 mgm.), but it assimilated only a small amount of nitrogen. The decomposition of the fraction of the straw soluble in alkalies and precipitated by hydrochloric acid is of especial interest. This fraction comprises lignins and certain pentosans (the preparation not being boiled in acid, for these experiments). The Zygorhynchus and the bacterium practically did not attack this fraction at all. The Trichoderma decomposed about one-third of it; this action was due to the presence of pentosans which were brought down with the lignins in the hydrochloric acid precipitate of the alkaline extract of the organic matter. The actinomyces decomposed considerably less total organic matter than the Trichoderma but a larger part of the fraction soluble in alkalies and precipitated by

acids. This is a result of the fact that this organism is actually capable of decomposing lignins in nature, as will be shown in a later contribution.

As a result of these and other experiments, the Trichoderma was selected for the following investigation:

A series of 10-gm. portions of rye straw were treated in a manner outlined in table 2. All were extracted first with benzene and then, after the removal of the benzene, with 95 per cent alcohol. Two portions of the straw thus treated were left as preparation 2. The other portions were treated, after the removal of the alcohol, with cold water for 24 hours, then filtered and dried. Two portions were again set aside, as preparation 3. Several of the remaining portions were treated according to the method of Willstätter (14) so as to obtain lignin directly. The other portions were extracted, at 15 pounds pressure for 30 minutes, with 50-cc. portions of 5 per cent sodium hydroxide solution, filtered, washed with water, then with dilute acetic acid, again with water, and then dried. Two of the portions thus treated were set aside as preparation 4. The remaining portions were boiled with 200 cc. of 2 per cent sulfuric acid solution for 2 hours, then filtered and washed, to give preparation 5. The lignin prepared according to the method of Willstätter formed preparation 6. The alkaline extract of the straw was neutralized with hydrochloric extract; the precipitate was filtered, washed with water and dried, to give preparation 7; it consisted partly of lignin and partly of pentosans. The untreated straw formed preparation 1. Sixteen 100-gm. portions of a Sassafras sandy soil and 16 portions of washed white sand were placed in 300-cc. long-necked flasks. Two-gram portions of preparations 1 to 5 and 1.5-gm. portions of 6 and 7 were added to the soil and sand flasks, leaving two flasks in each series as controls. Twenty-five-cubic centimeter portions of the first nutrient medium were added to all the flasks, which were then plugged and sterilized. The soil flasks were inoculated with a crude culture of cellulose-decomposing bacteria, containing also *Bacterium* No. 7 and protozoa, but free from fungi and actinomyces. The sand flasks were inoculated with a pure culture of the *Trichoderma*. The flasks were then connected with the respirator and incubated at 25 to 30°C. for 31 days. At the end of the period of incubation the residual ammonia and "humus" were determined; the latter was measured by extracting the soil or sand culture twice with sodium hydroxide, then precipitating the extract with hydrochloric acid. The "humus" given in table 5 represents the sum of the α-fraction and the organic matter of the β-fraction.

The results obtained are instructive. They show first of all that pure lignin is not utilized either by strong cellulose-decomposing bacteria or by fungi. When the lignin prepared by the Willstätter method was used, there was actually no trace of decomposition; when that part of the alkali extract which is precipitated with hot hydrochloric acid was used, some decomposition took place because of the presence of pentosans in the preparation.

The results further show that the lignin is found in the same preparation, obtained by the same methods commonly used for determining "humus" and "humic acids;" the lignin which accumulates in the soil forms at least a part of this "humus" or "humic acids." Practically all the lignin introduced with the straw or with the variously treated straw preparations is obtained again almost quantitatively in the final "humus." Preparations 4 and 5 from which most of the lignin has been removed by treatment with sodium hydroxide gave in the soil and sand media a much smaller amount of additional "humus" than preparations 1 to 3 from which the lignin has not been removed. The fact that there is an actual increase in the "humus" above that part which can be accounted for by the lignin shows another phenomenon, which will be brought

TABLE 5

Decomposition of straw and its constituents by the soil population and by Trichoderma

PREPARATION NUMBER*	AMOUNT OF MATERIAL ADDED TO 100 GM. OF SOIL OR SAND	MEDIUM†	CO_2 LIBERATED AS C								NH_3-N LEFT‡	NH_3-N USED UP	"HUMUS" CONTENT OF SOIL	EXCESS "HUMUS" OVER CONTROL
			2 days	4 days	7 days	13 days	21 days	31 days	Total	Total—control				
	gm.										*mgm.*	*mgm.*	*mgm.*	*mgm.*
Control		Soil	8.43	7.45	5.49	18.71	13.72	16.36	70.16		49.8		2,488	
1	2	Soil	13.82	16.37	23.42	61.74	46.45	37.30	199.10	128.94	29.1	20.7	2,967	479
2	2	Soil	20.68	18.33	28.02	37.35	26.45	40.28	171.12	100.96	28.8	21.0	2,989	499
3	2	Soil	14.11	14.61	17.54	54.70	29.20	37.30	167.46	97.30	30.9	18.9	2,756	368
4	2	Soil	12.05	12.05	11.00	31.28	58.90	42.06	167.34	97.18	35.4	14.4	2,712	224
5	2	Soil	15.97	8.82	12.54	33.12	32.06	47.04	149.55	79.39	34.2	15.6	2,719	231
6	1.5	Soil	6.86	5.19		19.40	22.94	17.16	72.05	1.89	46.9	3.0	3,618	1,130
7	1.5	Soil	7.55	16.37	13.13	19.80	22.14	19.20	98.19	28.03	44.4	5.4	3,084	896
Control		Sand	1.28				4.70	12.84	18.82		45.2			0
1	2	Sand	29.01	41.06	18.92	20.28	14.12	13.52	136.91	118.09	29.4	15.8		346
2	2	Sand	21.27	33.61	22.84	22.05	12.84	12.25	124.86	106.04	28.5	16.7		364
3	2	Sand	21.96	50.18	29.01	10.88	33.32	17.45	162.80	141.98	25.8	19.4		392
4	2	Sand	17.06	28.71	42.92	24.80	41.75	18.23	173.47	154.65	19.5	25.7		135
5	2	Sand	9.70	12.35	32.15	63.42	29.40	20.78	167.80	148.98	23.4	21.8		150
6	1.5	Sand	3.04				5.00	5.20	13.24	−5.58	45.0	0.2		1,287
7	1.5	Sand	2.06	14.70	5.59		7.45	4.12	33.92	15.10	43.5	1.7		816

* The preparation of the straw constituents is given in table 2; no. 1 is straw itself; no. 2 is straw from which the benzene- and alcohol-soluble fractions have been removed; no. 3 is straw from which the benzene- and alcohol- and cold water-soluble fractions have been removed; no. 4 has also the NaOH-soluble portion (5 per cent NaOH solution used under pressure) removed; no. 5 has also the rest of the pentosans removed, by boiling for 2 hours with 2 per cent H_2SO_4; no. 6 is lignin prepared by Willstätter method; no. 7 is the precipitate obtained by neutralizing the NaOH extract with HCl, washing and drying.

† Soil medium: 100 gm. of dry soil with 25 cc. nutrient solution sterilized and inoculated with crude suspension of cellulose-decomposing and other bacteria and protozoa (no fungi or actinomycetes). Sand medium: 100 gm. of washed sand with 25 cc. of nutrient solution sterilized and inoculated with *Trichoderma*.

‡ Control soil contains 4.6 mgm. NH_3-N.

out in detail later, namely, that a part of the 'humus" is actually synthesized in the soil by the action of microörganisms. In other words, the lignin contributes a part of the "humus" (not only quantitatively but also qualitatively) and the synthesized cells of the microörganisms contribute another part. The synthesizing action of the microörganisms is clearly brought out by the quantities of ammonia used up and converted into microbial protoplasm.

It is interesting to compare the action of the bacteria in the soil and of the fungus in the sand upon the different straw preparations. The strong cellulose-decomposing fungus makes a better growth, as indicated by the increase in CO_2 evolution and the ammonia consumption, with an increase in cellulose and pentosan content of the preparation (4 and 5). The bacteria, however, thrive better on the alcohol-, water-, and alkali-soluble extractives, although some of the celluloses and pentosans are also decomposed. This is in conformity with studies reported elsewhere (12) that the celluloses in normal humid soils are largely decomposed by the filamentous fungi of the soil.

Without going into a detailed discussion of the rate of decomposition of the various constituents of the straw, which is left for the following contribution where more extensive data are presented dealing with this subject, it is sufficient to point out the fact that both the evolution of CO_2 and the transformation of nitrogen from an inorganic into an organic form point to a much greater decomposition of the celluloses by the fungus when the lignins have been removed. In other words, the lignin in the straw not only does not decompose but it even prevents to a certain extent the decomposition of the celluloses and hemicelluloses. If CO_2 can be looked upon as a true index of the respiration of an organism and if the same organism produces the same amount of CO_2 when different carbohydrates are used as sources of energy, untreated straw is decomposed considerably slower than the straw from which the fats and waxes, water-soluble and alkali-soluble portions have been removed. The ratio between the carbon liberated as CO_2 and the nitrogen assimilated is very narrow in the case of the pure fungus culture, being about 7.5 for the untreated straw and about 7.0 for the cellulose obtained from this straw. This fact serves to illustrate what extensive quantities of nitrogen are required by the organism when straw is plowed under. The undecomposed straw will serve to keep the soil at a point of nitrogen starvation as far as higher plants are concerned as long as there is an excess of available energy.

SUMMARY

Among the various ingredients of natural organic material, such as straw, the lignins are most resistant to the action of fungi and bacteria; their accumulation in the soil accounts for a large part of the soil "humus," which is formed as a result of the decomposition of the straw. Since "humus" is usually considered as that part of the soil organic matter which is extracted by alkalies and precipitated by acids, the lignins added to the soil form one of the constituents of this "humus." This is due to the fact that most of the fungi and

bacteria which attack the natural organic substances added to the soil are unable to decompose lignins to any considerable extent; the lignins are thus allowed to accumulate in the soil, in the absence of organisms which are capable of decomposing them. The nature of these organisms will be discussed in detail later.

REFERENCES

(1) BENGTSSON, N. 1925 Bestämning av Inkrustered cellulosa i jord. Meddel. 279, Centralanst. Försöksv. Jordbruksområdet (Sweden). Bakt. Avd. 37.

(2) CHARPENTIER, C. A. G. 1921 Studien über den Einflusz des Rindvieh-und Pferdestallmistes auf die Zersetzung der Zellulose in der Ackererde. Diss. Helsingfors.

(3) COLLISON, R. C. 1925 The presence of certain organic compounds in plants and their relation to the growth of other plants. *Jour. Amer. Soc. Agron.* 17: 58–68.

(4) DORE, W. H. 1920 The proximate analysis of hardwood: Studies on *Quercus agrifolia*. *Jour. Indus. Engin. Chem.*, 12: 984.

(5) DORYLAND, C. J. T. 1916 The influence of energy material upon the relation of soil microörganisms to soluble plant-food. N. Dak. Agr. Exp. Sta. Bul. 116.

(6) FRAPS, G. S., AND HAMMER, N. C. 1910 Studies on the ammonia-soluble organic matter of the soil. Tex. Agr. Exp. Sta. Bul. 129.

(7) GORTNER, R. A. 1916 The organic matter of the soil. I. Some data on humus, humus carbon, and humus nitrogen. *Soil Sci.* 2: 395–442.

(8) HEUKELEKIAN, H., AND WAKSMAN, S. A. 1925 Carbon and nitrogen transformations in the decomposition of cellulose by filamentous fungi. *Jour. Biol. Chem.*, 66: 323–342.

(9) HUTCHINSON, H. B., AND CLAYTON, J. 1919 On the decomposition of cellulose by an aerobic organism (*Spirochaeta cytophaga*, n. sp.). *Jour. Agr. Sci.* 9: 143–173.

(10) JOHNSEN, B., AND HOVEY, R. W. 1918 The determination of cellulose in wood. *Jour. Soc. Chem. Indus.* 37: 132T–137T.

(11) WAKSMAN, S. A., AND LOMANITZ, S. 1925 Contribution to the chemistry of decomposition of proteins and amino acids by various groups of microörganisms. *Jour. Agr. Res.* 30: 263–281.

(12) WAKSMAN, S. A., AND SKINNER, C. E. 1926 Microörganisms concerned in the decomposition of celluloses in the soil. *Jour. Bact.* 12: 57–84.

(13) WAKSMAN, S. A., AND STARKEY, R. L. Microbiological analysis of soil as an index of soil fertility. VII. Carbon dioxide evolution. *Soil Sci.* 17: 141–161.

(14) WILLSTÄTTER, R., AND ZEICHMEISTER, L. 1913 Zur Kenntnis der Hydrolyse von Cellulose. I. *Ber. Deut. Chem. Gesell.* 46: 2401–2412.